RESISTANCE

FUNGICIDE RESISTANCE ASSESSMENT

National Agricultural Pesticide Impact Assessment Program (NAPIAP)

FUNGICIDE RESISTANCE

IN THE

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PREFACE

Resistance to systemic fungicides in the United States is present and increasing as the use of these chemicals proliferates. Although there are many fungicides registered for use in the U.S., the phenomenon of cross-resistance prescribes that, from the standpoint of fungicide resistance there are a very limited number of choices based on fungicide groups with common modes of action.

The broad spectrum, multi-site protectant fungicides, such as the EBDC's, chlorothalonil and captan, have not had resistance problems and are vital in the management of resistant pathogens. Disease management requires the greatest possible chemical diversity in fungicide options.

In view of the potential regulatory activity relating to the multi-site inhibitors fungicides, an assessment of fungicide resistance in the United States was made. Patricia L. Sanders, The Pennsylvania State University, through a grant from the Cooperative State Research Service's National Agricultural Pesticide Impact Assessment Program, undertook this study.

A panel of experts was convened on March 7-8, 1989, to discuss the current status of resistance to systemic fungicides in the United States. Included were representatives from U.S.D.A. and E.P.A., American representatives of companies that have fungicides in development or in market in the U.S., and once academic expert in each of the following crop areas: apples, citrus, fruits and nuts, grapes, ornamentals, peanuts, potatoes, tobacco, turf, and vegetables. Following are individual summaries of discussion in these crop areas, plus summaries of the discussion on fungicide resistance in strawberry production and mushroom production. Also included is a summary of the discussion on fungicide resistance as it relates to EPA policies and priorities.

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EXECUTIVE SUMMARY

Resistance to fungicides in pathogen populations is probably the most significant problem confronting modern U.S. agriculture in the area of chemical disease management. At present, there are available for broad spectrum control of plant pathogens only three groups of systemic fungicides: the benzimidazoles, the dicarboximides, and the ergosterol biosynthesis inhibitors. Cross-resistance is common within these groups, and although there is a plethora of commercially-available fungicides, the phenomenon of cross-resistance prescribes that there are, in effect, only three broad-spectrum systemic fungicides available. Any pathogen population that is resistant to one fungicide within a group will probably, by cross-resistance, be insensitive to other members of that group. The same situation exists in the phenylamide group of systemic fungicides which control Oomycete pathogens.

If manufacturers of fungicides were continuously developing new disease control chemistries, the foregoing situation would be less serious. However, the discovery and development of novel disease-active chemicals is increasingly costly and time consuming. This assures that there will be fewer new compounds forthcoming, and that those which are synthesized will be slower to the marketplace. Since fungicides are likely to continue as the mainstay of disease management for the foreseeable future, effective control of plant diseases requires effective monitoring, prevention, and management of fungicide resistance in pathogen populations to insure the longest possible useful life of fungicides and avoidance of major crop losses from chemical control failures. Equally essential is the preservation of the greatest possible chemical diversity in fungicide options through maintaining the availability of the multi-site protectant fungicides.

Commercial introduction of the first single-site inhibitors, the benzimidazoles,was followed worldwide by reports of control failures attributable to selections by these fungicides of highly resistant pathogen populations. Similarly, there have been serious and rapidly-developing control failures and crop losses with the phenylamide fungicides outside the U.S. Likewise, control failures with the dicarboximide fungicides have occurred in Europe in greenhouse and field, although these have been more slow to develop in use situations. With the sterol biosynthesis inhibitors, there are reports of control reduction and evidence of increasing levels and frequency of resistance in Europe to these fungicides in populations of Erysiphe graminis on cereal crops, Sphaerotheca fuliginea on cucumbers, and Erysiphe cichoracearum on melons.

Many of the control failures with the dicarboximide, phenylamide, and sterol biosynthesis-inhibiting fungicides have occurred in Europe. Systemic fungicides in these three groups have originated from European manufacturers and the chemicals, therefore, have longer use histories in Europe. These fungicides, however, are coming into increasing use in U.S. agriculture, and the experience in Europe with these fungicides indicates that problems of resistance in target pathogen populations will undoubtedly ensue.

It is clear that fungicides are likely to continue as the mainstay of disease management strategies for the foreseeable future. (Sanders) Effective control of plant diseases requires monitoring, prevention, and management of fungicide resistance in pathogen populations. This is necessary to ensure the longest possible useful life of the fungicide and avoidance of major crop losses from chemical control failures. Equally essential is the preservation of the greatest possible chemical diversity in fungicide options by maintaining the availability of multi-site protectant fungicides.

This assessment provides information on the current status of resistance to systemic fungicides in the United States for the following crop areas: apples, citrus, fruits and nuts, grapes, mushrooms, ornamentals, peanuts, potatoes, strawberries, tobacco, turf, and vegetables. In addition, EPA priorities and policies related to fungicide resistance are discussed. conclusions and recommendations are provided at the end of the report.

I. FUNGICIDE RESISTANCE IN APPLE PRODUCTION

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Introduction

The apple disease spectrum varies from region to region, but can include as many as ten or more fungi. Annual severity of individual diseases within a region is related to weather patterns in a given year or over a period of years. The grower's choice of fungicides is based on the desired control spectrum, fungicidal action required (protectant or curative), cost, compatibility with other fungicides and insecticides, potential for phytotoxicity (especially on fruit), and availability. Disease management strategy may be related to cultivar susceptibility, but most current commercial cultivars are, at least, partially susceptible, and some fungicides are required for disease management on all cultivars in all regions. The grower prefers to have a suitable spray mix for all the cultivars in an orchard block. In the US, apple disease management practices have been affected by fungicide resistance in scab (*Venturia inaequalis*) and blue mold (*Penicillium expansum*).

Apple Scab

Fungicides and Resistance Incidence

Dodine - Resistance has been reported in many areas of Michigan and western New York, one confirmed location in VA, and occasional grower complaints in PA. Still effective in many areas.

Benzimidazoles (benomyl, thiophanate-methyl) - Confirmed resistance has been reported in most eastern regions. Not recommended for scab control in MI and NC. Useful in mixtures in some areas of MD, NY, OH, PA, VA and WV.

Resistance to both the benzimidazoles and dodine has been confirmed in some orchards in MI and one orchard in VA. No scab resistance is suspected in major acreage areas of WA, but resistance to dodine and benzimidazoles has been confirmed in British Columbia, about 50 miles north of WA.

SBI's (fenarimol, myclobutanil, triforine - currently registered) - No resistance has been reported in the U.S. Stanis and Jones (7) reported reduced sensitivity in field isolates from West Germany. In an article now in press (2) Hildebrand, et al., associate loss of control by bitertanol with reduced sensitivity of the fungus in Nova Scotia. Some resistant isolates have minimal, inhibitory concentrations (MIC) more than 20 times higher than a sensitive isolate. Varying degrees of cross-resistance are present with all of eight SBI's tested, including bitertanol, myclobutanil, etaconazole, penconazole, triflumazole, flusilazole, pyrifenox and triforine.

Alternative scab fungicides - (mancozeb, metiram and captan) -Registration of three other alternatives, captafol, dichlone, and glyodin has been withdrawn. The supply of the less-preferred protectant fungicides, ferbam and thiram is questionable. Thiram is considered weaker than EBDC fungicides. Ferbam leaves a black spray residue and can be deleterious to fruit finish. There is increasing concern in the apple industry about adverse publicity regarding many of the available compounds needed to combine with benzimidazoles and SBI's.

Resistance monitoring procedures:

Dodine - spore germination or agar plating techniques with isolated fungus are suitable to determine MIC. Less than five-fold reduction in sensitivity and day-to-day variation may require repeated tests.

Benzimidazoles - Plating of viable conidia from 10-20 lesions on PDA containing 2.5-5 g benomyl/ml and comparing growth response on non-amended PDA in 24 hr generally gives reliable results. Fungus need not be isolated in pure culture, but contaminants may interfere with the growth pattern of *V. inaequalis*. Lack of percent inhibition of spore germination alone is not a reliable indicator of resistance.

SBI's - Inhibition of spore germination is not a reliable indicator of sensitivity. Agar plating techniques with isolated fungus are suitable to determine MIC. Repeated tests may be required, depending on the range of sensitivity detected.

Resistance management strategies:

Dodine - Used for scab control only and limited to early season use.

Benzimidazoles - Recommended only in mixtures with protectants since first reports of resistance were known in 1975. Must be combined with effective fungicides for rust control in the Hudson Valley and mid-Atlantic region. Benzimidazole usage in WA orchards is discouraged to reduce potential for resistance in post harvest rots.

A study involving orchard trees inoculated with benomyl-sensitive and benomyl-resistant strains of *V. inaequalis* and treated with mixtures of different concentrations of benomyl and mancozeb (3) showed that an increase in the mancozeb concentration in the mixture tended to delay the buildup of the benomyl-resistant population.

SBI's - Grower usage patterns are not yet stabilized.

Blue Mold (Penicillium expansum)

Benzimidazoles (benomyl, thiabendazole, thiophanate-methyl) - Benzimidazoles have been applied to fruit entering storage or being packed for shipment. Resistance is common in eastern apple regions. Resistance can be readily monitored by agar plating techniques. Not all isolates obtained from air-sampling are highly pathogenic on fruit.

In NY, Rosenberger (5) demonstrated negatively correlated cross-resistance between the benzimidazoles and diphenylamine (DPA), an antiscald agent. Combinations of these fungicides control isolates highly resistant to benzimidazoles or DPA. Packing house and storage sanitation are important in preventing decay caused by a low percentage of isolates intermediate in resistance to benzimidazole/DPA treatments.

In the mid-Atlantic region, captan is often included with a benzimidazole fungicide in the antiscald drench treatment of fruit entering storage. Although not particularly effective for rot control, captan prevents the build-up of viable benzimidazole-resistant spores in the drench tank and broadens the control spectrum. The possible alternatives, DPA and chlorine, have potential for injuring some fruit cultivars. Imazilil is now registered for packing line application, but not for pre-storage drench treatment.

Use of benzimidazole fungicides in Washington orchards is discouraged by packing houses to maintain effectiveness of benzimidazoles applied during the packing operation.

The above information was compiled after personal communication with the following fruit pathologists February-March, 1989: R.P. Covey, WA; M.A. Ellis, OH; F.F. Hendrix, GA; K.D. Hickey, PA; A.L. Jones, MI; D.A. Rosenberger, eastern NY; W.H. Shaffer, MO; R.A. Spotts, OR; T.B. Sutton, NC; W.F. Wilcox, western NY; and E.I. Zehr, SC.

Withdrawal of EBDC fungicide registration for apples September, 1989 leaves only captan, thiram, ziram and ferbam as protectant fungicide alternatives for consideration in resistance management strategies. Besides the negative characteristics of these compounds listed above under alternative scab fungicides, the following aspects should be noted:

> Incompatibility of captan with spray oil prevents its usage in one or two early season applications if oil is used for insect management. Captan prices have rises sharply since 1989.
>
> The dithiocarbamates thiram, ferbam, and ziram are less effective and more expensive than were the EBDC's for control of many apple diseases, rendering them less desirable substitutes for EBDC's for resistance management purposes.
>
> Dodine is still available and may be used for scab control in one or two early season applications where resistance has not been detected in the mid-Atlantic region.

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II. FUNGICIDE RESISTANCE IN CITRUS PRODUCTION

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MAJOR DISEASES

1. GREASY SPOT (Mycosphaerella citri)

This disease, which causes severe leaf fall and fruit blemish in Florida, is initiated by ascospores released from fallen leaves in June-July. Benomyl was adopted by growers in 1974 because one spray during the period of spore discharge gave good control due to the eradicant action of this fungicide against incipient infections (10, 21). Benomyl-resistant strains of M. citri were reported in 1974 (18) following application of the fungicide once each year for 5 years. Ascospores of the resistant strains germinated normally on agar medium containing 100 ppm benomyl, and the resistant strains could not be controlled by recommended benomyl spray treatment.

Monitoring for the presence of resistance is not considered a feasible strategy because the resistant strains are widely dispersed, and the leaf litter in each grove would have to be monitored each year prior to ascospore discharge. Also, spores can move from grove to grove.

While oil alone may provide adequate control of greasy spot on citrus foliage, and possibly orange fruit, copper plus oil, a single application in July, is required for good control of greasy spot on grapefruit rind. Copper may accentuate rind scarring of grapefruit, however.

2. SCAB (Elsinoe fawcettii)

Conidia of the pathogen infect young growth (foliage and fruit). The fruit remains susceptible for about 3 months after petal fall, but the most serious impact on fruit quality results from early infection of the fruit (20,21). Benomyl was introduced in 1974 as a highly effective treatment applied once at 2/3 petal fall under low disease pressure with a second spray applied prebloom if disease pressure is high. By 1980, 25-100% of E. fawcettii isolates showed resistance to benomyl (19).

Monitoring for resistance is difficult because the pathogen is slow-growing and pustules of the pathogen are contaminated with bacteria and other fungi making it difficult to routinely assay the resistance of isolates on benomyl-amended medium. Whiteside (19) determined resistance of E. fawcettii isolated by macerating pustules of the pathogen and applying the conidia and hyphal fragments to the apeces of a rough lemon that had been root-treated with benomyl solutions.

In the absence of benomyl-resistant strains of _E. fawcettii_, a single spray of benomyl applied at 2/3 petal fall should give effective disease control when followed by 1-2 sprays of copper. When benomyl-resistant _E. fawcettii_ are present, copper should be applied 2-3 times to the fruit surface at 2-3 week intervals beginning at 2/3 petal fall, because the copper treatment has purely protective action. Captafol applied late dormant and at 3/4 petal fall, was a highly effective replacement for benomyl but is no longer registered for citrus.

3. GREEN MOLD (_Penicillium digitatum_ and _P. italicum_)

The intensive and continuous use of biphenyl, sodium _o_-phenylphenate (SOPP), thiabendazole, benomyl, and _sec_-butylamine to prevent decay of citrus fruits after harvest has resulted in a serious problem of fungicide-resistance in _Penicillium digitatum_ and _P. italicum_. Serious losses of the crop have been encountered in all citrus producing areas of the world (Table 1). (6). Penicillium isolates that are resistant to one fungicide are usually cross-resistant to structurally-related compounds, e.g., biphenyl and sodium _o_-phenylphenate; thiabendazole and benomyl (Fig. 1). Treatment of fruit with one fungicide before storage usually results in the buildup of resistant isolates in the pathogen population so that a structurally-related fungicide cannot be used effectively on this same lot of fruit after storage (Fig. 2).

The section pressure exerted by a postharvest fungicide treatment on citrus fruits is considerably greater than that produced by a spray application of the same fungicide in the field. The postharvest fungicide is applied in a manner that ensures complete coverage and the fungicide deposit decreases only slightly during the storage life of the fruit. The treated fruit are stored or "ripened" under conditions that permit growth and sporulation of fungicide-resistant biotypes of the pathogen. The number of propagules (conidia) of _P. digitatum_ can increase about 10^8-fold in 7 days under optimum environmental conditions. These fungicide-resistant conidia are dispersed readily by air currents in the packinghouses to other fungicide-treated fruit which continues the process of selection for the fungicide-resistant biotypes. Commercial fruit handling practices that encourage the development of a practical problem of fungicide resistance in _Penicillium_ decay of citrus fruits have been discussed in detail elsewhere (1,6,7,10,11; Table 2).

Practical problems of fungicide resistance have not been reported in other post-harvest diseases of citrus fruits. _Diplodia natalensis_ and _Phomopsis citri_, which cause stem-end rots of citrus fruits, have been subjected to considerable selection pressure by application of benzimidazole fungicides in citrus groves, and fungicide-resistant isolates have been reported (17). Apparently, resistance has not become a post-harvest problem because dispersal of inoculum from the pycnidia is limited compared to _Penicillium_, and there is little opportunity for the disease to spread after harvest. Isolates of _Diplodia natalensis_ that were resistant to biphenyl were described 30 years ago (6). Other diseases caused by _Geotrichum_, _Trichoderma_, _Alternaria_ and _Botrytis_ have

not been effectively controlled by selective fungicides, so the presence of resistant strains would pass unnoticed.

The management of the resistance problem in Penicillium is currently an area of active research. The layout and management of most packinghouses is sufficiently unique that procedures to control resistance efficiently must be customized to specific circumstances (1,11). Two approaches dominate in the development of an overall strategy (Table 3; Fig. 4): 1) Sanitation measures and antisporulant treatments to suppress the spore population to a very low level; 2) Fungicide programs that do not encourage the proliferation of fungicide-resistant strains in the spore population. The principal methods for reducing the spore inoculum level in a packinghouse are: i)isolate and eliminate decayed fruit and the spore-ladened atmosphere from the packinghouse; ii) disinfest spore-contaminated fruit by spraying them with a hypochlorite solution before they are conveyed into the packinghouse, and disinfest the fruit-handling equipment and premises with an appropriate biocide; iii) if possible, treat fruit before storage with an effective antisporulant such as imazalil; iv) recondition cartons of deteriorating fruit from storage, when required, outside of the packinghouse. The importance of sanitation and anti-sporulant measures is evident from the fact that the number of fungicide-resistant spores in the fruit environment is proportional to the total spore population. When the total population is reduced substantially through sanitation, fewer mutants will be available for selection by a fungicide treatment. Furthermore, a small spore population in the environment will reduce the likelihood that a critical mass (inoculum threshold) of fungicide-resistant spores will be exceeded at an injured site on the surface of a fruit treated with the selecting fungicide (23).

During periods of the year when fruit are not harvested, packinghouses can be disinfested with broad-spectrum antimicrobial agents (e.g., formaldehyde, quarternary ammonium compounds) that cannot be used when fruit are present. This practice assures a very low population of spores in the packinghouse at the beginning of the harvest period, resulting in high efficiency of all postharvest fungicide treatments.

A study of the benefits of an inoculum-reduction (sanitation) program was carried out in three California lemon packinghouses (1,11). Packinghouse layout and sanitation significantly influenced the size of the population of thiabendazole-resistant spores and the level of decay in lemons shipped to distant markets. Effective strategies involved the isolation of sources of fungicide-resistant strains, disinfestation of contaminated equipment and containers, and a program for monitoring the level of fungicide-resistance in the packinghouse. An economic analysis of fruit quality and prices received at the market showed that the strategies for controlling fungicide-resistant Penicillium were economically justified (11).

Application of several fungicides in an appropriate sequence can prevent, or at least delay, the buildup of fungicide-resistant strains

of _Penicillium_ (Fig. 3; Fig. 4). This strategy is especially valuable in situations where the fruit are treated with fungicides both before and after storage or a holding period (e.g., degreening, lemon storage). The fungicides applied before storage are chosen on the basis that they will not encourage the proliferation of _Penicillium_ strains during storage that are resistant to the fungicides applied after storage. The latter treatments are critical since they must protect the fruit from decay during shipment and marketing. For example, lemons can be treated with fungicides chosen from sodium carbonate, _sec_-butylamine, potassium sorbate, borax, and imazalil before storage and, after storage, with SOPP, thiabendazole and biphenyl. The success of this strategy depends upon an efficient sanitation program because the buildup of a large population of fungicide-resistant strains in the packinghouse could jeopardize the post-storage fungicide treatments.

Over the past decade, several new fungicides that control benzimidazole-resistant strains of _Penicillium_ have been tested (2,3,9,10,12,14). Imazalil, prochloraz, and etaconazole belong to a class of compounds that inhibit the biosynthesis of ergosterol, an essential component of the fungus cell membrane. Generally, mutants "resistant" to these compounds show only a low level of tolerance and are often less virulent that the wild-type strains of the pathogen. Laville (16) streaked spores of fungicide-sensitive isolates of _Penicillium digitatum_ and _P. italicum_ once each week for 3 years onto agar plates containing benomyl or imazalil. He also subjected spores of these fungi to mutagenic agents (N-nitrosoguanidine and UV radiation). Many mutants appeared that were resistant to benomyl, but no imazalil-resistant mutants were found. DeWaard _et al_. (4) reported the isolation of _P. italicum_ strains, after UV irradiation of spores, that possessed elevated levels of resistance to imazalil. All of the isolates were pathogenic to oranges. The sensitive (wild) isolates were controlled by treatment of inoculated fruits with a solution of imazalil (250 mg/liter), whereas the resistant mutants were not well controlled at this dosage. Imazalil (500 mg/liter) controlled all of the _Penicillium_ isolates tested. There is considerable discussion today regarding the probability of development of serious resistance to imazalil and other fungicides with a similar mode of action. Nonetheless, biotypes of _Penicillium digitatum_ were found recently in California packinghouses which are not controlled by commercial treatments of imazalil (8).

Guazatine, a broad-spectrum di-guanide compound, is very active against benzimidazole-resistant isolates of _Penicillium_ and the fungus _Geotrichum_ that causes sour rot of citrus fruits (3,7). Unfortunately, variants of _Penicillium_ that are resistant to guazatine have been isolated recently in Australia and New Zealand (15,22).

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Table 1. Estimated* effect of imazalil upon decay losses in California citrus fruits in the 1979 season

Variety	Current Control Measures		With Imazalil		Percent Decrease	
	Cartons	Dollars	Cartons	Dollars	Cartons	Dollars
Lemons	5,323,400	48,706,000	129,108	11,552,450	98	76
Oranges	4,424,000	30,998,000	1,659,000	11,613,000	63	63
Grapefruit	672,000	3,783,200	235,000	1,411,200	63	63
Tangerines	110,400	673,440	55,200	336,720	50	50
Total	10,484,800	$84,150,640	3,240,280	$24,913,370		

Table 2. Factors influencing fungicide resistance in *Penicillium* spp. on citrus fruits.

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1. Frequency of resistance biotypes in the grove.

    a) Field sprays

2. Fungicide selection pressure in the packinghouse: Intensity and duration.

    a) Non-selective treatments
    b) Mixtures, rotations, and dosage

3. Fruit condition and environmental factors influencing disease development.

4. Production, dissemination, and survival of spores.

    1) Antisporulants
    2) Packinghouse layout
    3) Sanitation

5. Relative fitness and other interactions of resistant and sensitive biotypes.

Table 3. Strategies for Management of Fungicide-resistant *Penicillium* in citrus packinghouses.

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1. Emphasize non-selective fungicides - heat, chlorine, sodium carbonate.
2. Alternate organic fungicides with different biochemical action - e.g., thiabendazole and imazalil.
3. Prevent dispersal of *Penicillium* spores.
4. Monitor spore population to detect buildup of fungicide-resistant biotypes.

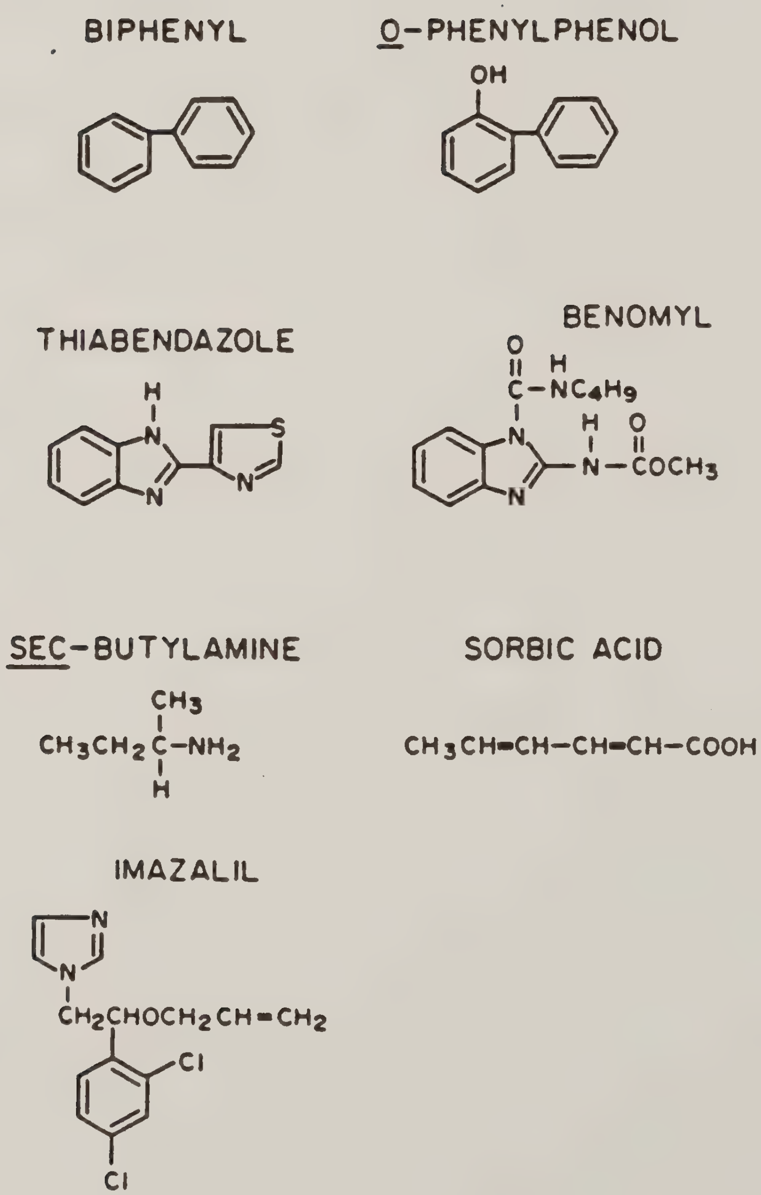

Figure 1. Fungicides applied in commercial packinghouses to control fruit decay.

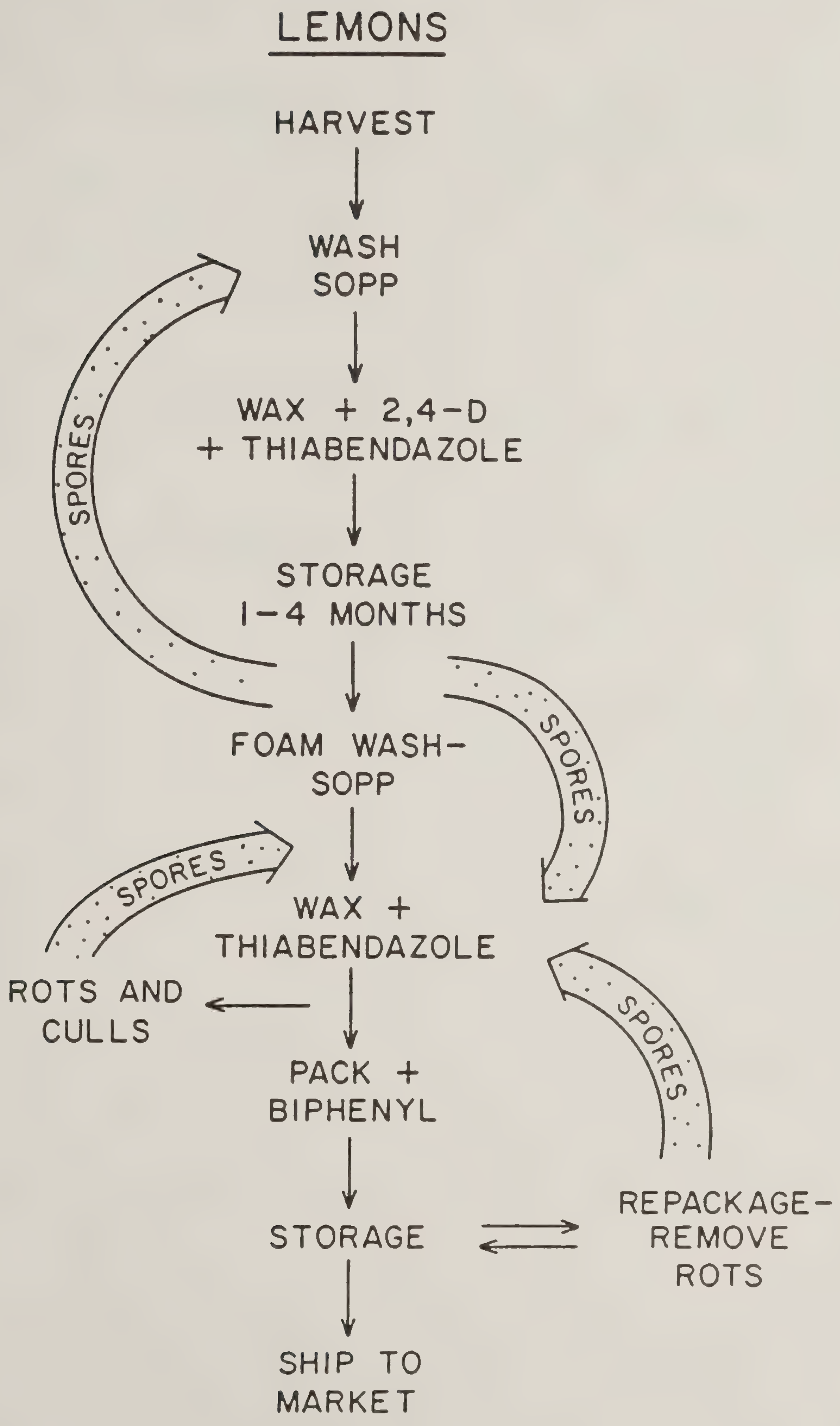

Figure 2. Flow diagram of fresh lemons in a typical California packinghouse showing fungicide treatments, storage at 15°C, and dissemination of fungicide resistant Penicillium spores

Fig. 3 A STRATEGY FOR MANAGEMENT OF FUNGICIDE-RESISTANT PENICILLIUM IN HARVESTED LEMONS

STAGE OF HANDLING	FUNGICIDE TREATMENT[1]
HARVEST ↓	
FUNGICIDE BATH ↓	3% SODIUM CARBONATE 41-43°C
WATER WAX + 2,4-D + FUNGICIDE ↓	IMAZALIL OR POTASSIUM SORBATE OR BORAX
STORE AT 12°C, 1-4 MONTHS ↓	
DUMP STORAGE BOXES (ISOLATED ROOM) ↓	HYPOCHLORITE SPRAY
WASH FOAM OR FLOOD ↓	SODIUM *O*-PHENYLPHENATE
WATER WAX + FUNGICIDE ↓	THIABENDAZOLE OR BENOMYL
PACK IN CARTONS ↓ (or → HOLDING ROOM)	BIPHENYL/PAPER

SHIP TO MARKET ← REMOVE DECAYED FRUIT (ISOLATED ROOM) ← HOLDING ROOM

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[1]Fungicides listed are registered in the U.S.: *Sec*-Butylamine, guazatine, and carbendazim can be applied in some other citrus-

III. FUNGICIDE RESISTANCE IN FRUIT AND NUT PRODUCTION

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INTRODUCTION

I. Production

In the United States, fruit and nut crops are commercially grown in most of the 50 states. The 1985 USDA[2] (USDA Table 252) figures show production acreages of major deciduous fruits (including pome fruits) as 1,758,200 acres, and nuts, including almonds, filberts, macadamias, and walnuts, as 655,700 acres. For later comparisons, acreage estimates and value (x 1000 dollars) of these crops are presented for states with the highest acreage or production figures.

Stone Fruits & Nut Crops	Acres	($Value X 1000)	State[1,2] (Cv[3])
Stone fruits			
Apricots	20,792	($50,989)	California (13)
Peaches & Nectarines	57,764	($224,668)	California (167)
Plums & Prunes	112,711	($302,819)	California (63)
Sour Cherries	44,000	($68,098)	Michigan (1)
Sweet Cherries	9,554	($101,485)	California (8)
Nut Crops			
Almonds	416,092	($725,697)	California (40)
Filberts	30,000	($16,738)	Oregon (10)
Macadamias	13,500	($30,450)	Hawaii (2)
Pecans	20,300	($166,288)	Georgia (12)
Pistachios	39,202	($56,736)	California (2)
Walnuts	171,708	($232,173)	California (15)

[1]California Farmer. 1988, California at a glance - Crop year 1987. HBJ Farm Publications, 731 Market Street, San Francisco, CA 94103-2011.

[2]USDA. Agricultural Statistics 1986. US Government Printing Office, Washington:1986. 551 pages.

[3]Number of major cultivars, including pollenizers.

Resistance management strategies used currently and those developed in the future will differ between states, within regions in the state, and even among neighboring adjacent orchards judging from the varied major production centers, number of states growing fruit and nut crops[1], and the large number of cultivars grown. Strategy differences are attributed primarily to the wide diversity of crops, differences in environmental conditions, cultural

practices and disease levels present. Cultivar differences and their susceptibility to diseases require specific management strategies to ensure effective disease control for each cultivar. Thus, problems of fungicide resistance in plant pathogens in the orchard must be discussed in terms of specific crops, number of diseases controlled by each fungicide application, and availability of management tools. To prevent or delay the development of resistance in pathogens to fungicides, research critical to developing management strategies are: 1) predicting resistance risk with each pathogen-fungicide combination, 2) monitoring procedures for early detection of resistance problems, based on comparisons using baseline sensitivities of pathogens before introduction of fungicides in the orchard, 3) evaluating population dynamics of resistant pathogens in competition with the fungicide-sensitive isolates, and 4) understanding the mechanisms of resistance in the pathogen.

II. Fungal diseases (pathogens)

Examples of major plant pathogens exposed to fungicides used for disease control on stone fruit and nut crops are listed below. Yet, at this time, resistance problems occur with only a few pathogens as indicated (*) and not in all regions.

Alternaria spot and fruit rot (*Alternaria alternata*)
Anthracnose (*Glomerella cingulata*)
Black knot (*Dibotryon morbosum*)
Brown rot blossom blight and fruit rot (**Monilinia laxa* and **M. fructicola*)
Ceratocystis canker (*Ceratocystis fimbriata*)
Cherry leaf spot (**Coccomyces hiemalis*)
Cladosporium rot (*Cladosporium herbarium*)
Cytospora canker (*Leucostoma persoonii*)
Eutypa dieback (*Eutypa armeniacae*)
Fusicoccum canker (*Fusicoccum amygdali*)
Gray mold rot (**Botrytis cinerea*)
Gilbertella rot (**Gilbertella persicaria*)
Green fruit rot (**Botrytis cinerea*, *Sclerotinia sclerotiorum*)
Leaf curl (*Taphrina deformans*)
Leaf blight (*Seimatosporium lichenicola*)
Leaf rust (*Tranzschelia discolor*)
Mucor rot (*Mucor piriformis*)
Peach tree short life (Complex of bacterial canker, Cytospora canker and/or cold injury)
Penicillium fruit rot (**Penicillium expansum*)
Phymatotrichum root rot (*Phymatotrichum omnivorum*)
Phytophthora crown and root rot (*Phytophthora* species)
Powdery mildew (*Sphaerotheca pannosa*, and *Podosphaeria clandestina*)
Rhizopus rot (**Rhizopus stolonifer*, **R. arrhizus*, *R. circinans*)
Rust (*Tranzschelia discolor*)
Russet scab of prune (Cause unidentified)
Scab (*Cladosporium carpophilum* on stone fruit and **C. carigenum* on pecan)
Shoestring crown and root rot (*Armillaria mellea*)

Shot hole (Stigmina carpophilia)
Verticillium wilt (Verticillium dahliae)

III. Fungicides recommended or tested for control of stone fruit and nut crop diseases.

A. Sulfur

1. Dust and wettable sulfur
2. Liquid sulfur (Lime and sodium polysulfide)
3. Gaseous sulfur (sulfur dioxide)

B. *Copper (tank mix and fixed)

1. Tank mix copper (Bordeaux mixture)
2. Fixed copper (Basic copper sulfate, carbonates, hydroxides, and oxychlorides)

C. Organic fungicides

1. Dithiocarbamates (thiram, ziram, ferbam, maneb, zineb, mancozeb, polyram, prothiocarb)
2. Quinone (dichlone)
3. Imidazolines (glyodin, captan, captafol, folpet)
4. Guanidine (dodine)
5. *Antibiotics (streptomycin, hydroxytetracycline, cycloheximide)
6. *Benzimidazoles (thiabendazole, benomyl, thiophanate methyl, carbendazim)
7. *Sterol biosynthesis inhibitors or demethylation inhibitors (triadimefon, flusilazole, bitertanol, imazalil, prochloraz, fenarimol, nuarimol, triforine, propiconazole)
8. *Dicarboximides (vinclozolin, iprodione)
9. Other fungicides
   a. Metalaxyl (Ridomil)
   b. Aluminum tris (o-ethyl-phosphonate) (Aliette)
   c. Tutane (Butafume)
   d. *Dicloran (Botran)
   e. Terrazole (Truban)
   f. Fenaminosulf (Dexon)
   g. Dinocap (Karathane)
   h. Quintozene (Terraclor)
   i. Fentin hydroxide (Du-ter)
   j. Hypochlorous acid (Clorox)

*Resistance of pathogen(s) reported for fungicide classes or groups under laboratory or field conditions.

IV. Present resistance status

The first evidence of resistance of a fruit pathogen to fungicides in the United States was in 1962 when control of Penicillium fruit decay was reduced on citrus fruit treated postharvest with sodium orthophenylphenate (SOPP) and stored for extended periods before marketing (Harding, 1962). At this time, fresh market stone fruits in CA were also being treated postharvest with SOPP to control Monilinia and Rhizopus decay, yet resistance problems were not experienced. The probable reason was its limited usage since peaches are not stored for extended periods as is common with citrus. In 1963, fungicide screening programs in CA and NJ showed that DCNA (2,6-dichloro-4-nitroaniline) was very effective in control of Rhizopus rot of stone fruits. During the same year (Ogawa, et al., 1963) was able to develop DCNA-resistant Rhizopus stolonifer in culture. DCNA use was generally restricted to postharvest treatments of peaches, plums, nectarines, sweet cherries, and apricots, and widescale field applications were not made. DCNA resistance problems have not been reported from postharvest applications on stone fruits.

The advent of the benzimidazole fungicides for commercial use in 1972 was shortly followed by resistance problems with certain pathogens: Monilinia fructicola (1976), Monilinia laxa (1973), Botrytis cinerea (1971), Penicillium expansum (1975), Fusicladium effusum (1976) and Cladosporium carpophilum (1978). Also, dodine resistance has been reported for Venturia inaequalis (1969); however, resistance of Cladosporium caryigenum has not been detected, even with multiple applications on pecans. More recently, dicarboximides (iprodione and vinclozolin) and the demethylation inhibitors (triforine and propiconazole) have been introduced. Problems of field resistance have not been detected in the U.S. but have been reported with vinclozolin in Monilinia in Australia and Botrytis on grapes in France.

V. Individual Crops, Diseases, and Resistance Problems

A. STONE FRUITS

1. APRICOTS (Prunus armeniaca L.) are grown in CA (97.3%), WA (1.8%), and UT (0.9%) with increasing acreage in AZ.

   a. Brown rot blossom blight and fruit rot, caused by Monilinia laxa and M. fructicola, require one to three fungicide applications (benzimidazoles, chlorothalonil, captan, dicarboximides, triforine, or copper) during bloom. Preharvest sprays are generally not applied. Resistance problems, with associated crop failures, have arisen only with multiple applications of benzimidazole fungicides during bloom. The disease can be controlled effectively with alternative fungicides that have little or no resistance risk. Monitoring has been conducted by growing the pathogen on potato dextrose agar amended with 1 to 10 g/ml (a.i.) benomyl or methyl benzimidazole carbamate.

b. Shot hole, caused by _Stigmina carpophila_, is also a disease requiring fungicide applications during the winter to protect dormant buds, and again during the spring to protect against fruit infections. Two or three spray applications of copper (dormant), ziram or captan (after bloom) have been used for decades, but resistance problems resulting in crop failures have not been reported. Monitoring of field isolates for reduced sensitivity to copper, ziram or captan has not been conducted.

c. Green fruit rot is caused by _Botrytis cinerea_, _Monilinia_ spp. and _Sclerotinia sclerotiorum_. This complex of pathogens is controlled with the same blossom sprays used for brown rot control. Registered fungicides are benzimidazoles, captan, chlorothalonil, and dicarboximides. Resistance has not been reported for _Botrytis_ or _Sclerotinia_ on apricots to these fungicides.

d. Eutypa dieback is caused by _Eutypa armeniaca_ which attacks pruning wounds made during the wet winter months. Application of concentrated benzimidazole suspensions on pruning wounds as a protective treatment has not resulted in selection of benzimidazole-resistant strains of _Eutypa_ in the U.S.

2. PEACHES AND NECTARINES (_Prunus persica_ L. Batsch. and _P.persica_ var. _nectarina_ Maxim.) are produced in over 32 states with major production areas CA (68.5%, including 45.9% clingstones for processing and 22.6% freestones for fresh market), SC (10.7%), NJ (4.4%), GA (4.2%), PA (1.9%), WA (1.3%), and 9.0% in other states.

a. Brown rot blossom blight and fruit rot are caused by _Monilinia fructicola_, with some _M. laxa_.

In CA, control with fungicides during blossoming requires one to two spray applications of benzimidazoles, dicarboximides, demethylation inhibitor (triforine), chlorothalonil, captan, or coppers (bloom only), and two to three spray applications preharvest. Benzimidazole-resistant _Monilinia_ are now widespread in CA. Where the number of applications is lower (CO, TX), benzimidazole resistance problems have not been reported. Postharvest fungicides (spray and dip) are triforine, benzimidazoles, and DCNA.

In the eastern US, where brown rot blossom blight is a minor problem, chlorothalonil or captan is the first spray, applied during early bloom. A second spray is applied at the shuck-split stage for control of scab. Preharvest brown rot fungicides, sulfur, polyram, and mancozeb, are directed at scab control. If brown rot becomes a serious problem, dicarboximides or demethylation inhibitors are used. In orchards where multiple applications of benzimidazoles were

used, benomyl-resistant <u>Monilinia</u> has been reported (MI, NY, SC).

In summary, benzimidazole resistance problems, with associated crop failures, are widespread in the major peach producing areas of the US. Yet, in some states (AR, OR, TX) where benzimidazoles are used, benomyl-resistant <u>Monilinia</u> isolates have not been detected. In NJ, isolated cases of benomyl resistance were reported in 1985, but with the use of alternative fungicides, resistance was not detected in 1988. Detection of iprodione resistance with crop failures from brown rot has not been reported in the U.S.

A review of benzimidazole resistance shows that the first report of <u>M</u>. <u>fructicola</u> resistance to benomyl in the U.S. was a report by Jones and Ehret (1976) in MI, where multiple applications were practiced. Resistance levels ranged from 100 to 1,000 g/ml benomyl (Jones, 1983). In NY, Szkolnik and Gilpatrick (1977) reported that 9% of the benomyl-resistant isolated showed profuse mycelial growth in medium amended with 50 g/ml benomyl, while 63% showed profuse mycelial growth at 10 g/ml benomyl. In the summer of 1977, Ravetto detected isolates of <u>M</u>. <u>fructicola</u> resistant to benomyl in a peach orchard in CA where repeated applications of benomyl + captan combinations were made. This finding was confirmed by Ogawa and Manji, and the <u>M</u>. <u>fructicola</u> strains were found to be resistant to only 1 to 4 g/ml, indicating that the standard test of 10 g/ml suggested by the chemical company would not detect this resistance level and explained the crop failures that occurred. Benomyl resistance level has remained at 1-4 g/ml, although the frequency of resistant population has increased.

Based on population dynamics (Ogawa, Manji, Akaskaveg, & Michailides, 1988), management strategies must include a rapid monitoring system to determine population ratios of sensitive to resistant isolates, as well as levels of benomyl resistance. Monitoring techniques used are variations of screening isolations on medium amended with benomyl or methyl benzimidazole carbamate (MBC). Such testing does not provide growers with information on frequency of resistant strains in the population. Thus, until a rapid technique of monitoring is available, benzimidazoles are applied until disease control failures are experienced. To prevent the development of resistant fungal populations, minimum number of applications of benzimidazole fungicides are commonly recommended, with alternation favored over mixtures of benomyl with contact fungicides. Mixtures of benomyl with a contact fungicide are used largely because of the label requirement established by the manufacturer. Thus, the common practice of growers is to use fungicides (dicarboximides and demethylation inhibitors) rather than benzimidazoles, where resistance problems have been

experienced. Another possible strategy is the use of fungicides with negative cross-resistance to benomyl-resistant *Monilinia* sp.; however, such compounds, which have been tested in Europe, have not been available for testing in the US.

Management program used in California

Table 1. Examples of a management program used to control brown rot in fresh market peaches and nectarines in an orchard with high disease pressure.

I. Orchard with no resistant isolates, where brown rot has been controlled with benzimidazoles.

Recommendation: Continue to used present practice

Pink bud:	benzimidazole 1X
Preharvest:	benzimidazole 1X
Postharvest:	mixture of DCNA and benzimidazole

II. Orchard with low populations of resistant isolates, and low disease incidence of both blossom blight and fruit rot.

Recommendation: Reduce number of benzimidazole applications

Pink bud:	benzimidazole 1X
Full bloom:	dicarboximide, DMI, or chlorothalonil 1X
Preharvest:	Alternation of dicarboximide and DMI 2-5X
Postharvest:	mixture of DCNA, triforine, and benzimidazoles 1X

III. Orchard with high populations of resistant isolates, and either low or high disease incidence.

Recommendation: Omit use of benzimidazole applications

Pink bud:	dicarboximide 1X
Full bloom:	dicarboximide or DMI 1X
Preharvest:	alternate dicarboximide and DMI 2-5X
Postharvest:	mixture of DCNA and triforine 1X spray

b. Scab, caused by Cladosporium carpophilum is a serious disease in the eastern U.S. requiring multiple fungicide applications. Fungicide application begins at shuck split and continues throughout the summer for as many as 10 applications. Both benzimidazole and dodine resistance have been detected in some states but not in other states with the same multiple spray programs. Where benzimidazole and dodine resistance have been detected, the most commonly used fungicides are sulfur, chlorothalonil, polyram, and captan. Information on monitoring procedures was not found.

c. Shot hole, caused by Stigmina carpophilia, and leafcurl, caused by Taphrina deformans, require dormant spray of copper or

ziram, and more recently, chlorothalonil is registered for use in CA. Although one to two sprays of copper or ziram have been applied, resistance or control failures have not been reported. In the eastern U.S., shot hole and leaf curl are not a serious problem as it is in the western states.

d. Powdery mildew causal agents, Sphaerotheca pannosa and Podosphaera leucotricha are important pathogens on fruit in the western U.S. Sulfurs and benzimidazoles are applied, but resistance problems have not been evaluated because crop failures have not been observed.

3. PLUMS AND PRUNES (Prunus salicina Lindl. and P. domestica L.) for fresh market are grown in CA (76.6%), OR (11.5%), MI (11.0%), WA (4.7%), and ID (2.1%) and prunes for drying are grown exclusively in CA.

a. Brown rot blossom blight and fruit rot is caused by Monilinia fructicola with some M. laxa. The disease and its management is discussed under peaches.

b. Green fruit rot caused by Botrytis cinerea, is discussed under almonds.

c. Postharvest diseases, caused by a complex of fungal organisms, are discussed under peaches.

d. Black knot, caused by Dibotryon morbosum is a serious disease only in the eastern U.S. For effective disease control, six applications of benzimidazole plus captan are required. Multiple applications of benzimidazoles have not resulted in selection of resistant D. morbosum.

4. SWEET AND SOUR CHERRIES (Prunus avium L. and P. cerasus). Sweet cherries are produced in WA (28.5%), MI (23.3%), OR (21.8%), CA (17.8%) with other states (MI, ID, NY, PA) contributing 8.7%, while sour cherries are produced in MI (76.8%), NY (7.9%), UT (7.3%), with PA, OR, WI, and CO supplying the remainder.

a. Brown rot blossom blight, caused by M. fructicola and M. laxa, is important in most production areas, except in WA. Blossoms and fruit are highly susceptible and require 1-2 fungicide treatments during bloom. One to three preharvest sprays are also applied, especially on sweet cherries. Most commonly used fungicides are benzimidazoles and dicarboximides. The demethylation inhibitor, triforine, is used in orchards without Botrytis problems. With multiple benomyl applications during bloom and preharvest, benomyl resistant Monilinia and Botrytis have developed causing crop failures. In CA, use of benomyl has been withdrawn by the manufacturer. Thiophanate-m is still recommended for use in CA. Resistance problems have not emerged with the use of iprodione in the U.S. Monitoring technique is the same as that used on peaches.

b. Green fruit rot is caused primarily by *Botrytis cinerea* and *Monilinia* spp., and blossom sprays are most important to prevent quiescent infections. With benzimidazole-resistant *Botrytis* and the withdrawing of benomyl use on sweet cherries in CA, present treatments are iprodione or chlorothalonil, with some growers still using thiophanate-m without apparent failures.

c. Powdery mildew, caused by *Podosphaera oxycanthae* (*Podosphaera tridactyla*), affects both the foliage and fruit. Multiple spray applications of sulfur from bloom to harvest have provided control. More recently, triforine has been shown to be an excellent alternative. Resistance problems have not been reported.

d. Cherry leaf spot, caused by *Coccomyces heimalis* has been the most important disease on sour cherry. Benomyl and dodine are the recommended fungicides. There is some question of resistance to both benomyl and dodine in MI. Leaf spot has not been a major problem in OR or WA.

B. NUT CROPS

1. ALMONDS (*Prunus dulcis*) are produced exclusively in CA.

a. Brown rot blossom blight caused primarily by *Monilinia laxa* with scattered instances of *M. fructicola*, requires 1-2 annual applications of fungicide (benzimidazoles, dicarboximides, triforine, or captan) for disease management. Benzimidazole-resistant *M. laxa* has been detected in several orchards in CA. In these orchards, resistant populations have reached 60-100% and degrees of resistance 1-4 g/ml. Resistance has been monitored by placing mycelial agar plugs onto potato dextrose agar amended with benomyl or methyl benzimidazole carbamate. Where resistance problems developed, multiple applications of benzimidazoles had been made, alone or in combination with captan. In most orchards, only a single spray has been applied at the pink bud stage of bloom and in these orchards benzimidazole resistance problems have not been reported.

b. Green fruit rot, caused by *Botrytis cinerea*, *M. laxa*, and *Sclerotinia sclerotiorum* has been sporadic. Effective control has been obtained with a single benzimidazole application at early bloom with no detection of resistant isolates of the pathogens. Under high disease pressure in orchards without a history of benomyl-resistance, alternation of benzimidazole and dicarboximide fungicides during bloom would seem appropriate. Method for monitoring *Botrytis cinerea* is the same as that for monitoring *Monilinia* where appropriate fungicides are incorporated into potato-dextrose agar medium.

c. Shot hole disease, caused by *Stigmina carpophila* and scab, caused by *C. carpophilum* require 1-3 applications during bloom

to 5 weeks after petal fall. Chemicals used are ziram, captan, and iprodione. Ziram and captan have been used for many years with no indications of disease control failures. Iprodione was introduced recently, requiring 2-3 applications without alternation or use of mixtures. Resistance in Stigmina or Cladosporium to fungicides has not been reported for ziram, captan, or iprodione.

2. CHESTNUTS (Castanea dentata)

   a. Chestnut blight, caused by Endothia parasitica is the limiting factor in production of chestnuts in the U.S. Fungicides, including the benzimidazoles and 8-hydroxyquinoline, have been tested experimentally but not used commercially. Biological control methods are under investigation.

3. FILBERTS (Corylus avellana) are produced almost exclusively in OR (98.7%), with a few acres in WA (1.3%).

   a. Perennial canker, caused by Anisogramma anomala, is the most serious disease problem in OR, as the disease continues to spread without an effective control program. Two copper sprays are the only means of control, and resistance to coppers based on failures to control the disease has not been detected. Alternative control measures are needed judging from the poor performance of copper fungicides.

4. MACADAMIAS (Macademia ternifolia) are grown exclusively in HI.

   a. Phytophthora raceme blight, caused by Phytophthora capsici is the most important disease and is currently controlled with fosetyl-aluminum sanctioned by an Emergency Use Permit (EUP). Resistance problems have not been detected. Acreage increases in other regions of HI could result in increased disease problems. Although selection of fosetyl-aluminum resistance in Phytophthora has not been reported, alternative nonrelated fungicides should be introduced to make alternation available in the management strategy.

   b. Botrytis blossom blight, caused by Botrytis cinerea, is a minor problem, which was effectively controlled with captafol (registration cancelled). Now only the benzimidazoles are available for control. Because the disease is only of minor importance, benzimidazole fungicides are not used, except in few instances. Disease incidence could become more important as acreages expand, and production areas become located where B. cinerea populations are higher. In such a circumstance, benzimidazole applications should continue to be restricted to a single spray or alternated with another fungicide, such as the dicarboximides. Research should continue on an alternative fungicide for registration in the event Botrytis blossom blight becomes a serious problem.

5. PECANS (Carya illinoensis) are produced in GA (34%), TX (31.9%), NM (11.9%), AL (6.5%), LA (6.1%), OK (4.1%), MI (2.7%), FL (1.1%), AZ (0.7%), SC (0.6%), and NC (0.4%).

   a. Scab, caused by Cladosporium caryigenum requires multiple fungicide applications during the season. Registration and exclusive use of a benzimidazole fungicide resulted in selection of resistant isolates, and its use has been severely curtailed with these recommendations: (1) Do not use benomyl or thiophanate-m in consecutive applications for leaf control, (2) Do not use benomyl or thiophanate-m alone, (3) Where a second spray application is needed, use one of the tin fungicides. Triphenyltin hydroxide, dodine, and propiconazole are now used on an alternating schedule with no failures or resistance to this compound. Management of resistance is based on three applications of propiconazole, one pre-pollination followed by two cover sprays at 2 week intervals followed by three cover sprays of triphenyltin hydroxide or dodine at 2-3 week intervals. Orchards with multiple applications of triphenyltin hydroxide should be monitored for selection of resistance before crop failures occur, based on the fact that resistance of Cercospora beticola to phentin hydroxide was noted on sugar beets (Giannopolitis, 1978). In SC, 7-12 applications are normal and up to 18 applications under severe disease pressure. In GA, the fungicide schedule recommends pre-pollination spray application every 14 days from bud break through pollination and post-pollination spray every 2-3 weeks as needed before early August. After August, fungicides are applied as needed based on thorough orchard scouting.

      Research in GA (Reilly - personal communication) showed that elimination of late summer sprays (August) did not significantly affect scab control, but the lack of protectant fungicides at that time resulted in emergence of two other diseases (Phytophthora cactorum, infection of shuck and kernels and Glomerella cingulata, infection of the shuck) causing poor filling of kernels. Late summer sprays are again being considered, and a request for an EUP registration for fosetyl-aluminum fungicide is being considered for Phytophthora control. Benomyl and ziram are being considered for reducing the Glomerella problem. Other diseases such as zonate leaf spot, brown spot, and liver spot, which are now considered minor, could become major problems with reduction in use of protectant fungicides. Management programs should carefully monitor disease incidence and pathogen resistance before crop failures are experienced.

   b. Downy spot, caused by Mycospharella caryigena, attacks only the foliage. Spray applications should begin at bud break followed by cover sprays at 3-week intervals throughout the summer with such fungicides as benomyl, triphenyltin hydroxide (TPTH),

propiconazole, and dodine. Resistance problems have not been reported.

c. Powdery mildew, caused by Microsphaera alni, affects both foliage and nuts. Benomyl and sulfur are applied every 3-4 weeks for control. Resistance problems have not been reported.

d. Zonate leaf spot, caused by Cristulariella pyramidalis, attacks mature leaflets. Benzimidazoles are recommended in a combination treatment with TPTH or propiconazole. Resistance problems have not been reported.

6. PISTACHIOS (Pistacia vera L.) are produced primarily in CA, with some acreage in AZ.

a. Botrytis blossom and shoot blight is caused by Botrytis cinerea, while Botryosphaeria panicle and twig blight is caused by Botryosphaeria spp. Both diseases occur sporadically in CA, but in some orchards, crop losses reach epidemic proportions. Current control measure requires one application of a benzimidazole fungicide during bloom. Resistance problems and failure of disease control have not occurred. Yet, if multiple fungicide applications are necessary to control Botryosphaeria panicle and twig blight, resistance could be a problem. Because multiple benzimidazole applications can result in selection of resistant isolates, chlorothalonil and copper fungicides have been selected as alternates for future registration.

VI. Future resistance likelihood

The management of fungicide resistance on fruit and nut crops in the U.S. depends on the implementation of known strategies as well as development of new ones to prevent or delay resistance:

1. Availability and application of fungicides with different modes of action decreases the selection of fungicide-resistant isolates. Selection of resistant isolates occurs with exclusive, multiple application of at-risk fungicides.

2. Development and registration of fungicides with lower risk of resistance selection (resistance to dicarboximides and demethylation inhibitors has been slower to develop under field use situations).

3. Development of fruit cultivars with greater disease resistance. Such cultivars should reduce the number of fungicide applications required for disease control.

4. Development of biological control measures as alternatives to reduce use of fungicides.

5. Crop production in areas that are less disease-prone, where minimal chemical control is necessary.

6. Requirement for chemical companies to monitor resistance levels and frequencies of fungicide-resistant pathogens. This type of monitoring effort can detect possible failures in fungicide efficacy before major crop losses are experienced.

7. Fungicide use protocols based on research data will reduce the chance for selection of fungicide-resistant strains.

8. Provide research on the epidemiology and adaptation of resistant isolates under field conditions, and determine why resistance has not been a problem in certain regions.

9. Identify timing schedules for fungicides, which decrease selection of resistant populations. Application of fungicides should be precisely timed through resistance monitoring and/or disease forecasting systems.

10. Goals of near-perfect disease control should be lowered and the public educated to accept lower quality standards in crops.

11. Rapid assay methods to monitor pathogen populations for resistant isolates in order to develop early warning systems.

12. Establish centers and/or containment facilities for testing fungicide-resistant pathogens from different localities without fear of spread to crop production areas.

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IV. FUNGICIDE RESISTANCE IN GRAPE PRODUCTION

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I. MAJOR DISEASES

A. BOTRYTIS BUNCH ROT - *Botrytis cinerea*

1. Chemicals Used

a. Multisite inhibitors
captan
dithane M45

b. Benzimidazoles
benomyl
1) First reported field control failure (US)
Pearson, R.C., et al. 1979. Benomyl-resistant strains of *Botrytis cinerea* on apples, beans, and grapes. Plant Disease 64:316-318.

In NY, benomyl was not aimed primarily at *Botrytis* on apples, beans, and grapes. Thus, the development of benomyl-resistance in *Botrytis* populations may be classified as development in nontarget organisms. This situation was predicted by Ogawa et al. in 1976.

The use of tank mixes of unrelated fungicides has been recommended to avoid buildup or resistant strains. Authors state, however, that resistant isolates occurred even though captan was used in the tank mix, but no attempt was made to investigate what the population frequencies might have been under a tank mix vs. no mix program.

The results obtained by Pearson et al. suggest that the presence of benomyl-resistant strains of *B. cinerea* may not in all cases be explained by the history of benomyl usage. This implies and is supported by results of other research that some level of resistance existed in native populations prior to benomyl introduction. *Botrytis cinerea*, having an exceptionally wide host range ranging from weeds and vegetables to tree crops with no known strain development in pathogenesis, has been implicated in the spread of resistance among crops by spread of airborne conidia.

2) Confirmed field control failures

California - Sall, M.A. 1982. Unpublished.

Northover, J., and Matteoni, J.A. 1986. Resistance of *Botrytis cinerea* to benomyl and iprodione in vineyards and greenhouses after exposure to the fungicides alone or mixed with captan. Plant Disease 70:398-402.

c. Dicarboximides
iprodione (Rovral)
vinclozolin (Ronilan)
dicloran (Botran)
1) First reported field control failure
Northover and Matteoni. 1986. Canada - see reference above.
2) Confirmed field control failures
None in the U.S.

2. No Resistance Monitoring Procedures in Use

3. Resistance Management Strategies

a. Widespread resistance to benzimidazole fungicides has required reduced use and/or tank mixtures with multisite inhibitors such as Dithane M45 or Captan. No research basis in U.S.

Sall, M.A. Unpublished. Control correlated with captan use.

b. Use of Dicarboximides restricted to 3-4 applications. No research basis.

c. Studies in CA and Switzerland have shown that pruning to reduce foliar canopy reduces disease and allows more complete penetration of fungicide spray. Although more study is needed, canopy reduction may reduce or obviate the need for fungicides to control Botrytis bunch rot.

4. Population Dynamics

a. In vineyards, captan used in mixtures with either benomyl or iprodione for one year gave protection against Botrytis bunch rot of grapes, but it did not prevent an increase in the incidence of *B. cinerea* resistant to benomyl or iprodione.

b. Sall, M.A. 1982. California. Use of benomyl or benomyl + captan resulted in increased levels of benomyl-resistant *B. cinerea*. In all cases, the use of captan did not prevent the occurrence of benomyl resistance in the *B. cinerea* population.

B. POWDERY MILDEW - *Uncinula necator*

1. Chemicals Used

a. multisite inhibitors
sulfur

b. benzimidazoles
benomyl
1) First reported field control failure
Pearson and Taschenburg. 1980. Benomyl-resistant strains of *Uncinula necator* on grapes. Plant Disease 64:677-680.

Benomyl was used extensively in NY on grapes to control powdery mildew especially since sulfur is phytotoxic to Concord. In addition, growers used benomyl to control oxidant stipple, an air pollution problem that is widespread in grape-growing areas of the Great Lakes region. The development of benomyl resistance in the U. necator populations created problems of significant magnitude, because alternative fungicides were either much less effective (folpet) or were phytotoxic (sulfur, copper, lime and dinocap).

c. DMI's
triadimefon (Bayleton)
fenarimol (Rubigan)

1) First reported field control failure
Gubler, et al. 1990. Sensitivity of *Uncinula necator* populations in California to triadimefon. In preparation.

Reduced sensitivity to DMI fungicides on grapes in North America was unknown in all production areas until 1985, three years after the introduction of triadimefon into the U.S. In 1985, failures of triadimefon in grape powdery mildew occurred in a few commercial vineyards in California as well as in University of California research trials. In 1986, similar control problems again occurred but to a much great extent. Control was difficult to achieve, even in the Central Valley production areas. While it is true that DMI's exhibit a directional shift type resistance which in normal years would not be manifested as large scale crop failure, in 1986, weather conditions were conducive for rapid population buildup, and it is suspected that this, in conjunction with poor coverage and stretch spray intervals, allowed a population to increase which was made up of isolates with reduced sensitivity to triadimefon.

In the fall of 1986, investigations into the possibility of a resistance problem in California were begun. After several months of trying to develop screening techniques that would give reproducible results, a technique using a detached leaf test as modified by Pearson, et al. was used. Results of tests revealed that all isolates could be controlled by true protectant application of label rates of triadimefon. However, at rates below label there existed significant variation in sensitivity to triadimefon within the population of *U. necator* from vineyards having control problems.

Eradication of *U. necator* with triadimefon also was investigated. Results of these studies showed that while one isolate which was never exposed to triadimefon was effectively eradicated from grape leaves up to 7 days after inoculation, another isolate could not be controlled when spraying was done only 4 days after inoculation. This indicates that real differences exist in sensitivity to triadimefon among isolates.

2. Suspected control failure

In a trial of fungicides for control of powdery mildew on Concord grapes in 1988 in northwestern PA, disease incidence on triadimefon-sprayed grapes was not significantly different from non-treated vines. Several other fungicides provided excellent control (J.W. Travis, personal communication).

3. <u>Resistance Monitoring Procedures in Use</u>

   a. None for benomyl.

   b. Gubler (unpublished). Plans are to continue monitoring populations in selected vineyards in California. In addition, baseline sensitivity data are being collected for fenarimol and myclobutanil--25 vineyards and 30 isolates per vineyard.

4. <u>Resistance Management Strategies</u>

   a. Reduced use of benomyl in NY and use of DMI's.

   b. In California, use of DMI's only after early season applications of wettable sulfur. In addition, in vineyards where control with triadimefon was not achieved, switch to fenarimol or myclobutanil and tank mix with wettable sulfur through bloom applications. No research basis.

5. <u>Population Dynamics</u>

   a. Field studies indicate that where benomyl-sensitive strains of *U. necator* predominated, control of powdery mildew by the reduced-rate fungicide combinations could be attributed solely to the benomyl component. On the other hand, where resistant isolates were not predominant, disease control with combinations of benomyl plus another fungicide, each at reduced rates, depended on the efficacy of the comparison product. Therefore, if the nonbenzimidazole component of the mix has low efficacy or shorter residual, even full rate combinations may not provide economic control. For example, benomyl + captan at full rates failed to control resistant strains of *Botrytis cinerea* in New York vineyards.

   b. Greenhouse investigations indicate isolates resistant to triadimefon may revert slowly back to wild type. No field data yet to support this preliminary data.

V. FUNGICIDE RESISTANCE IN ORNAMENTALS PRODUCTION

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Hundreds of different species of woody and herbaceous plants are being grown as ornamentals in field and greenhouse production. Chemicals used on these crops are registered for 'minor use,' because the name of the plant to be treated must be listed. Little of any particular chemical is used on any one crop. However, if the total amount used on the general group 'ORNAMENTALS' is calculated, that amount is not 'minor.'

A huge variety of plant material is grown under a wide range of conditions, exposed to many different populations of pathogens and insect vectors of pathogens, and then shipped throughout the U.S. Plant material is also shipped to and imported from Europe, South and Central America, the Far and Middle East, and Australia. If a population of pathogen develops resistance to a chemical, it is inevitable that the population will be sold and shipped to other producers and the consumer.

It is extremely important to the ornamental industry that chemicals which are not at risk or are at very low risk to resistance development be retained in the disease control arsenal. In particular, mancozeb and chlorothalonil are extremely important materials for use as alternatives to or in conjunction with systemics that are at risk to resistance development. Of major concern is the fact that the procedures for registering a chemical on ornamentals requires that the name of the particular crop to be treated must be listed on the label. Thus, the use will always be considered 'minor' on a crop by crop basis. As a direct result, the importance of a chemical to ornamentals may not be given full consideration when the cancellation of that chemical is being contemplated. For example, if the bulk of a given chemical is used on ornamentals in general but cancellation is based on its importance on potatoes as compared to geraniums (rather than ORNAMENTALS IN GENERAL), cancellation may not appear to have a significant impact. While geranium producers may not suffer, ornamentals producers as a group may be devastated by the loss.

<u>Major diseases</u>

These vary with the particular crop. The list of the pathogens or diseases where fungicide use is intense during production is as follows:

<u>Foliar:</u>
- Botrytis (all crops)
- Powdery mildew (roses)

<u>Roots and stem:</u>
- Pythium (all crops)
- Phytophthora (primarily woody plants)
- Rhizoctonia (all crops)
- Cylindrocladium (foliage plants)
- Thielaviopsis (some herbaceous and some woody plants)

In terms of fungicide resistance, species of Botrytis have been the pathogens of major concern.

Chemicals commonly used

Fungicides are usually applied in greenhouses to control particular pathogens. Broad spectrum activity is not a major objective. In outdoor production of woody ornamentals, broad spectrum of activity is sought when controlling foliar pathogens. However, the development of resistant fungal populations on woody ornamentals may not be a problem since isolated plants or very small populations of plants rather than large numbers of closely spaced plants in monoculture are being treated.

Benzimidazoles
- benomyl (Benlate, Benomyl, Tersan 1991)
- thiophanate methyl (one active ingredient in Zyban and Banrot)
- thiophanate ethyl (3336-WP, 3336-F)

Dicarboximides
- iprodione (Chipco 26019)
- vinclozolin (Ornalin)
- dicloran, DCNA (Botran)

Dithiocarbamates
- zineb (Zineb)
- mancozeb (Dithane F-45)

Sterol inhibitors
- triadimefon (triazole, Bayleton)
- fenarimol (pyrimidine, Rubigan)
- triforine (morpholine, Triforine, Funginex)
- dodemorph (morpholine, Milban)

Dinitrophenol
- dinocap (Karathane)

Substituted aromatic
- chlorothalonil (Exotherm Termil, Daconil 2787, Bravo)

Phthalimide
- captan (Captan)

Acylalanine
- metalaxyl (Subdue, Ridomil)

Organic
- etridiazole (Truban, Terrazole, one a.i. in Banrot)

Metal-organic
- phosethyl-Al

Carbamate
propamocarb

Gray mold - Botrytis spp.

Resistance monitoring procedures in place

NONE

Resistance detection:

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Botrytis cinerea isolated from greenhouse crops in PA are being checked for resistance to benzimidazoles, dicarboximides, and chlorothalonil as they are received or collected from growers.

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Botrytis cinerea, *B. elliptica*, and *B. tulipae* from tulips, Easter lilies, and Asiatic lilies grown in the field or greenhouse are checked for resistance to benzimidazoles and dicarboximides upon request.

Resistance incidence

Botrytis cinerea from infected floricultural crops in PA greenhouses were examined in 1988 in vitro. (Gary W Moorman, The Pennsylvania State University)

5 populations resistant to benzimidazoles but not to dicarboximides
1 population resistant to chlorothalonil but not dicarboximides, or benzimidazoles
14 populations resistant to both dicarboximides and benzimidazoles

Those resistant to benzimidazole *in vitro* overcome the label rate of benomyl on excised plant tissue and sporulate. Some of those resistant to dicarboximide *in vitro* overcome the label rate of vinclozolin on excised plant tissue and sporulate while others do not. The chlorothalonil-resistant isolate does not overcome the label rate of chemical in vivo.

Decreased sensitivity was reported by growers but complete control failures have not been reported. It is suspected that the rigorous manipulation of temperature and relative humidity in greenhouses alleviated the Botrytis problem in PA. Without such environmental modification, control failures in operations relying on fungicides alone probably would occur frequently.

Gary A. Chastagner (Washington State University)

Botrytis cinerea, B. elliptica, and B. tulipae from tulips, Easter lilies, and Asiatic lilies grown in the field or greenhouse operations are checked for resistance at growers' requests. No ongoing monitoring is being conducted.

Lilies

Chastagner, G. A. and K. Riley. 1987. Phytopathology 77:1237 (Abstr.) Work was done to detect resistant populations in the state of Washington.

B. elliptica - 43% from lily fields and 87.3% from greenhouses were resistant to both benzimidazoles and dicarboximides.

B. cinerea - 8.3% from lily fields and 59.3% from greenhouse lilies were resistant to both benzimidazoles and dicarboximides.

B. elliptica developed resistance to dicarboximides after 2 years of use in the field and a control failure was documented. 80% of isolated obtained from the field were resistant (G. A. Chastagner, pers. communication).

Tulips

B. tulipae populations have resistance to benzimidazoles. While laboratory populations can be developed with resistance to dicarboximides, dicarboximide resistance is not a problem in fields.

Chastagner, G. A. and K. Riley. 1987. Phytopathology 77:1237 (Abstr.)

Monitoring was done for 6 years on outdoor grown tulips.

After 5 years of continued field use, no resistance to dicarboximide appeared. After 2 years, benzimidazole resistance was common. B. tulipae - 14% from fields and 100% from greenhouses resistant to benzimidazoles. Disease control failures in the field were reported to occur and the presence of resistant populations verified.

Resistance of Penicillium (blue mold) to benzimidazoles on bulbs is known, and field control failures have been documented.

Chrysanthemums

Webster, A. G. and C. E. Koons. 1973. Increased tolerance to benomyl in greenhouse populations of Botrytis cinerea. Phytopathology 63:1218-1219. Report of Botrytis on chrysanthemum resistant to benzimidazoles.

Resistance management strategies in place or recommended

In PA, current recommendations call for rigorous use of temperature and humidity manipulation in greenhouses for the control of Botrytis supplemented by the alternated application of chlorothalonil or mancozeb and

dicarboximides. Benzimidazoles will no longer be recommended for use in Botrytis control. However, the formulated mix with dithiocarbamate (Zyban) is still recommended.

Chastagner, G. A. 1984. Phytopathology 74:821 (Abstr.) In WA, it is recommended that dicarboximides not be applied to tulips until bloom but then can be used during bloom. Data indicate that either alternating or mixing full rates of mancozeb or chlorothalonil with dicarboximides on lily prevented the buildup of resistant populations in the field. For economic reasons, alternation is the standard recommendation.

Availability of research

Population dynamics

A technique for cycling populations on tissue has been reported--Moorman, G. W. 1988. Technique for cycling fungicide-resistant Botrytis cinerea populations on geraniums. Phytopathology 78:1531 (Abstr.).

Chastagner, G. A. and W. E. Vassey. 1979 Phytopathology 69:914. (Abstr.). Dicarboximide sensitive isolates were plated on dicarboximide-containing agar. Of 9 isolates tested, one contained resistant spores. The frequency of resistant spores in the population was estimated to be from 1.8 X 10-6 to 4 X 10-5. The resistant subculture grew at half the rate of the sensitive cultures, but sporulation, sclerotial formation, and virulence did not differ.

Root diseases - Pythium spp.

Resistance monitoring

NONE

Resistance incidence

Pythium species from infected plants in PA greenhouses were collected by Moorman (The Pennsylvania State University), the Department of Plant Pathology Disease Clinic, and the PA Department of Agriculture Clinic.

In vitro resistance:

Over 20 populations of various species have been tested for fungicide resistance.

1 population of _Pythium_ _aphanidermatum_ was found resistant to acylalanines, etridiazole, and propamocarb. The grower suspected a control failure in the greenhouse after using acylalanine and etridiazole. This population does not appear to be highly virulent in currently employed in vivo tests.

Resistance management strategies

None

Availability of research

None

Other cases of known resistance in pathogens of ornamentals:

Resistance of Verticillium that attacks deciduous trees to benzimidazoles used in tree injection for wilt control. McHugh, J. B. and L. R. Schreiber. 1984. Plant Disease 68:424-427.

VI. FUNGICIDE RESISTANCE IN PEANUT PRODUCTION

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Peanut (Arachis hypogaea L.) is a major agronomic crop in the southeastern United States. It is one of the most profitable crops in this region and has enabled many growers to survive in recent years of depressed farm economies. Production is concentrated in seven states (GA, AL, NC, TX, VA, OK, and FL) which harvest about 600,000 ha annually (20). These states can be divided into three basic regions as follows: (1) VA and NC (VA/NC), (2) GA, FL, and AL, i.e., the southeast, and (3) TX and OK. Some production problems such as diseases are common to all three regions, while others are unique to one or two regions.

Unfortunately, peanut is susceptible to numerous diseases (20). Many currently grown cultivars were developed for their agronomic traits and have little disease resistance. Although this is changing with the introduction of new fungicide treatments, annual losses to some diseases are high, while others are limited by fungicide treatments. The high cost of chemical controls which routinely are $100-250/ha is justified by the value of the crop. Such heavy reliance on fungicides for successful peanut production is risky. Environmental and regulatory concerns are resulting in fewer options for chemical control, and new fungicides seem more prone to resistance problems.

The latter is the topic of this paper and is of grave concern to the peanut industry. The first two sections will each deal with specific major diseases where resistance has been reported. The third section will introduce several additional diseases, the common denominator being that all are controlled by the sterol biosynthesis inhibitors (SBIs). These compounds have been used only experimentally in peanut and appear to hold tremendous potential. SBI resistance has not been reported in peanut pathogens but must certainly be recognized as a possibility based on experience with other fungi (21). Resistance monitoring and use strategies for SBIs will be discussed in this section. The fourth and final section will summarize the known and potential impact of fungicide resistance on peanut production.

PEANUT LEAFSPOTS

Early leafspot (Cercospora arachidicola Hori.) and late leafspot (Cercosporidium personatum (Berk. & Curt.) Deighton) are both serious diseases of peanut throughout the world (20). Yield losses of 50% and greater are common where these diseases are not controlled.

Although one peanut cultivar with some leafspot resistance has been released recently (10), fungicides are still used on virtually 100% of the U.S. crop with multiple applications (three to ten) per season. Numerous fungicides have been used, including fentin hydroxide, captafol, chlorothalonil, copper

hydroxide, mancozeb, maneb, sulfur and benomyl (23). The primary pathogen in the VA/NC region is C. arachidicola which is easier to control than C. personatum. A variety of the listed fungicides are used in that region. In the southeast, C. personatum is prevalent and chlorothalonil has proven to be the most effective labelled product. It is the only fungicide recommended for late leafspot control in GA and, despite intensive use, has shown no indications of resistance problems (4).

Benzimidazole fungicides were introduced for use on peanut in the late 60's and proved very effective for control of foliar peanut diseases. Benomyl was used widely but surveys as early as 1973 indicated that in vitro resistance was already developing in GA (14). This was a high level resistance but was not linked to crop loss in the field. Simultaneous studies in AL confirmed this in vitro resistance of both C. arachidicola and C. personatum and reported decreased field efficacy (8). Subsequently, there were numerous disease control failures in the southeast. Resistant isolates persisted in nature making the reintroduction of benomyl impractical (4,22).

Experience in other peanut growing regions such as VA and TX suggest that the consistent use of companion fungicides may prevent or at least delay benzimidazole resistance. In TX, benzimidazoles were usually applied with mancozeb, primarily to aid control of web blotch (Phoma arachidicola) and rust (Puccinia arachidis). As of 1978, Smith, et al. (24) reported no known cases of disease control failure in growers' fields, although experimental plots treated only with benomyl contained high populations of resistant isolates. Use of benzimidazoles has subsequently declined with most growers now using chlorothalonil. There has been a similar decline in benzimidazole use in VA, where decreased efficacy has been noted in recent years, but isolates have not been checked for sensitivity (P.M. Phipps, personal communication).

History indicates that benzimidazole resistance is a stable trait in nature, and both C. personatum and C. arachidicola appear to be no exceptions (22). A 1987 survey found one C. personatum population with high benzimidazole resistance out of 13 randomly selected in GA (4). Currently, this poses no real hardship on the peanut industry due to the availability of highly effective, economical fungicide alternatives. Therefore, there is little activity concerning resistance strategies or monitoring. It is unlikely, but conceivable, that this could change if other alternatives are lost and new chemicals are not introduced to replace them.

SCLEROTINIA BLIGHT

Sclerotinia blight is a soilborne disease of peanut caused by the fungus Sclerotinia minor (Jagger) Kohn. Although not reported in the U.S. until 1971 (17), by 1982, it was considered the most important peanut disease in VA and OK (20). It consistently causes major yield losses in NC and TX as well but has not been a problem in the southeastern growing area.

Two cultivars with some resistance to Sclerotinia blight are available (1) but are not widely grown and losses continue to be high. Fungicides are needed to control this disease and allow even marginal profits from infested fields. PCNB provides some suppression of symptoms and dicloran, iprodione, and

vinclozolin have shown activity in field trials (5). Full registration was granted iprodione for use on peanut in 1985 and is still being sought for vinclozolin.

The in vitro sensitivity of S. minor to PCNB, dicloran, iprodione, and vinclozolin was found to be 1.27, 0.91, 0.11, and 0.07 g/ml, respectively (5). In vitro resistance to these fungicides was soon found (5,18,19). Resistance occurred at frequencies of 1.8 to 2.3% in the populations evaluated and resistant strains were capable of growth on media amended with 1,000 g/ml fungicide (5,18). They also exhibited cross-resistance to dicloran, iprodione, vinclozolin, PCNB, and procymidone (6,19).

This was cause for concern in light of the dicarboximide resistance reported in other fungi (15). A monitoring protocol was established which utilized media amended with 2 g/ml of iprodione or vinclozolin to identify resistant isolates. Numerous research and grower sites were monitored over a 3-year period with 763 isolates evaluated, none of which were resistant (5). Subsequently, one isolate was found with in vivo resistance that originated in a microplot receiving sequential treatments of iprodione (7). When additional peanut microplots were infested with this isolate and treated with fungicides, no significant decrease in control of disease occurred during a 3-yr period (26). It is thought that resistance is low level and not easily measured in terms of disease control or yield response, and there have been no losses linked to dicarboximide-resistant isolates of S. minor at this time.

The potential for development of dicarboximide resistance in the field is still a possibility. Further research with the in vitro resistant isolates showed them to be as virulent as their fungicide-sensitive 'parent' isolates and able to survive and compete in vitro (6). This is unlike dicarboximide-resistant isolates of Botrytis cinerea which often have decreased vigor and revert to their sensitive state when no longer exposed to the fungicide (16). However, the resistant S. minor strains still responded to fungicide applications in the field and were controlled just as well as the highly sensitive isolates. This parallels the early experience with dicarboximide-resistant Botrytis (16), but later work has shown that such isolates can be linked to reduce disease control (11). Studies with mixed populations of Sclerotinia isolates indicate that fungicide treatments selectively favor survival of resistant isolates (6). The impact of these selected isolates on the future of peanut production appears to be minor, but monitoring programs are still needed to detect any changes in the resistance situation.

Current resistance management strategies consist of minimizing the use of dicarboximide fungicides and monitoring treated populations. The former is achieved by stressing nonchemical controls (1) and scouting to apply these fungicides only when needed. Grower compliance has been good, in part due to the high cost of treatment.

Monitoring of treated areas is still being conducted by evaluating mycelial growth on fungicide-amended agar. An alternative technique utilizing fungicide-treated peanut stems inoculated with S. minor has also been developed (3). This method appears to be a better indicator of isolate response to dicarboximide treatment in the field than the method using

fungicide-amended growth medium.

Unfortunately, no effective alternative fungicides are currently available for resistance management. Some experimental compounds evaluated recently have given excellent control of Sclerotinia blight and have demonstrated activity against dicarboximide-resistant isolates (25). The future of these chemicals is not known, but their development and registration would offer a valuable management tool for combating Sclerotinia blight and any future development of field resistance to the dicarboximides.

DISEASES CONTROLLED BY SBIs

The SBIs offer control of a spectrum of peanut pathogens previously unknown in a single fungicide class. Originally evaluated for control of peanut leafspots, they soon were found to have activity on other diseases including southern stem rot (*Sclerotium rolfsii*) and Rhizoctonia limb rot (*Rhizoctonia solani* Kuhn (anastomosis group (AG) 4). There are numerous reports documenting this in recent issues of Fungicide and Nematicide Tests. At least one active compound, diniconazole (9), also demonstrated significant plant growth regulating properties, a desirable trait considering the current decline in use of daminozide (13).

The combined benefits of these fungicides are often reflected in large increases of peanut pod yields. Although no SBIs are currently labeled for use on peanut in this country, registration packages for several have been submitted to EPA. If these products are labeled, it is anticipated that they will receive widespread acceptance by peanut growers. This is particularly true of GA, FL, and AL where SBIs control three of the most damaging diseases present, (i.e., leafspot, southern stem rot and Rhizoctonia limb rot). Unfortunately, it is in these southeastern states where disease pressure and fungicide usage is often highest, thus compounding the risk of resistance.

Although resistance to SBIs has not been reported in peanut pathogens, it must be considered a possibility. There are two approaches currently employed to manage potential SBI resistance in peanut. First, survey work is being conducted to establish baseline sensitivities of pathogens from different geographic areas before these compounds are labeled and widely used. *C. personatum* is thought to be a high-risk pathogen due to its biology and past experience with benzimidazoles. A 1987 study conducted at the University of Georgia evaluated bulk samples from 13 locations in south GA and found them to have a high, uniform level of SBI sensitivity (4). In 1988, Mobay researchers looked at single-lesion isolates from six locations in Georgia and Florida. They evaluated germ tube growth on water agar amended with 0.01, 0.1, and 1 g/ml tebuconazole (Folicur 1.2 EC). Significant seasonal variation in sensitivities of *C. personatum* isolates was detected, and there was some evidence of population shifts toward decreased sensitivity where SBI's had been used at high rates. Monitoring will continue in 1989 (M.R. Schwarz, Personal Communication). Previous work with seven isolates of *C. arachidicola* and one isolate of *C. personatum* found EC_{50} levels for propiconazole to be 0.10 and 0.15 g/ml, respectively (12).

Less effort has been made to quantify the SBI sensitivity of soilborne pathogens such as R. solani (AG-4) and S. rolfsii. One study evaluated the in vitro sensitivity of 28 R. solani and Rhizoctonia-like isolates to diniconazole, cyproconazole, tebuconazole, PCNB and chlorothalonil. The mean EC_{50}'s for these fungicides were found to be 0.028, 0.056 0.166, 4.06, and 4.85 g/ml, respectively (2). Further work is needed to establish baseline sensitivities for all major peanut pathogens by sampling a wide variety of pre-exposure populations.

The second approach to SBI resistance management involves the design of use patterns to maximize the benefits of this chemistry for the grower while minimizing the resistance risk. As mentioned previously, chlorothalonil is used throughout the U.S. for foliar disease control and should provide an excellent companion product for the SBIs. The largest actual disease-related yield losses are from soilborne pathogens due to the inadequate control measures currently available. Since labelled fungicides provide good, economical foliar disease control, the biggest contribution of the SBIs should be their application for soilborne diseases. They can be applied on a broadcast basis or as a foliar spray and still be efficacious on these soilborne pathogens. Therefore, one scenario would be to apply SBIs only when needed for stem rot, limb rot, etc., and substitute them for the usual foliar fungicides, thus making them more price competitive. Another scenario would be to promote full season use of prepack mixtures or tank mixtures. There are positive and negative aspects to both cases from a theoretical point of view (27). In reality, sales considerations, marketing agreements, etc. will play a large role in decision making. The important fact is that good companion products are available and specific use recommendations will have to result from cooperation between industry and academia.

SUMMARY

In the light of the heavy use of fungicides on peanut, the crop has had surprisingly few problems from resistance. Certainly, the loss of the benzimidazoles was significant and cost the growers a valuable tool for foliar disease management. The impact of this has been reduced by the continued availability of other effective products.

Dicarboximide resistance is a concern, but at this time has not been a problem in the field. The future will tell us the significance of this phenomenon.

Efforts are being made to develop peanut cultivars with genetic resistance to our major pathogens, but it is a slow process. Fungicides will play an integral role in disease management for years to come and will be essential for profitable production. The benefits of these products are too great to risk losing them to resistance.

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VII. FUNGICIDE RESISTANCE IN POTATO PRODUCTION

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Major Diseases

A. Early blight caused by Alternaria solani.

B. Late blight caused by Phytophthora infestans.

Other fungal diseases are of relatively minor importance with respect to fungicide use.

There are currently no fungicide resistance problems in the U.S. with either pathogen. The potential problem with metalaxyl resistance in P. infestans is significant, and most of the remainder of this report applies exclusively to this potential problem. There is some potential for A. solani to develop resistance to dicarboximides. However, these chemicals are not currently used extensively and no resistance problems have been reported in A. solani (although dicarboximide resistance has been reported for other Alternaria species).

Commonly used fungicides

dithiocarbamates
chlorothalonil
triphenyltin hydroxide
fixed copper
dicarboximides (early blight only)
phenylamides (late blight only)

Metalaxyl is the only phenylamide currently registered in the U.S., although cross-resistance occurs among all chemicals in the class.

Monitoring

There are no systematic monitoring programs in place in the U.S. Ciba-Geigy, the manufacturer of metalaxyl, monitors resistance to metalaxyl only when samples are sent from outbreaks believed to be associated with any potential resistance problems (personal communication, Eileen King-Watson).

The most appropriate resistance assay is an in vivo test (e.g., the floating leaf disk test). Bulk samples may be appropriate for detecting low frequencies of resistance in a population but are not adequate for quantitative estimates.

Resistance incidence

Severe crop losses due to metalaxyl resistance in *P. infestans* were first reported from Europe in 1980 (Davidse, et al. 1983). Since then resistance has been reported in numerous countries throughout the world, including Mexico. No resistance has been found in the U.S.

Approximately 90% of the isolates in a sample of *P. infestans* taken in August, 1988 (W.E. Fry, unpublished data) from Saltillo, Mexico (near Monterey), less than 150 miles from the Rio Grande Valley, appeared to be metalaxyl-resistant. These isolates were tested using an amended agar, radial growth test; they will be tested further using a floating leaf disk test since radial growth studies are not conclusive. The suggestion here is that resistance may be knocking at our southern border, and it may be only a matter of time before metalaxyl-resistant individuals of *P. infestans* are found in the U.S.

There are at least three possible reasons why there is currently no metalaxyl resistance problem in *P. infestans* in the U.S.:

1. Resistant inoculum is not present.

   a. Not imported in infected seed.

   b. Has not migrated from Mexico yet.

   c. Has not mutated to resistance due to good disease control maintaining small pathogen populations (thereby reducing the probability of a mutation occurring).

2. Selection is not strong enough.

   a. Large amounts of metalaxyl may not have been used in most areas (especially because of the need to control early blight with broad-spectrum fungicides that also control *P. infestans*).

   b. In most potato production areas, the climate is not generally conducive to late blight, therefore, resistance problems have not developed rapidly.

3. Resistance evolves, then goes extinct.

   Resistant mutants may have occurred but could have gone extinct due to the fact that most inoculum (resistant or not) does not survive *in situ*.

Resistance management

The following recommendations have been made for foliar use of phenylamides in general (Urech, 1988, Urech and Staub, 1985):

1. Pre-packed mixtures with companion fungicide

2. High rates of companion

3. Maximum application interval of 14 days

4. Limit to 2-4 applications per crop, per season

5. Preventative (pre-infection), not post-infection use

6. No soil applications

7. Use all possible integrated disease control tactics

Additional recommendations specific for metalaxyl resistance management in _P. infestans_ in potatoes might include (Milgroom, unpublished):

1. Extra efforts at sanitation (cull piles, volunteers).

2. Use certified seed only

The theoretical rationale for these strategies can be summarized as follows (see Milgroom and Fry, 1988):

1. _Reduce growth rates of both resistant and sensitive types_

   a. Mixtures

   b. High rate of companion

   c. Application interval 14 days

   d. Integrated control

2. _Reduce differential selection_

   a. Limited number of sprays

   b. Foliar application only

3. _Keep initial frequency low or zero_

   a. Preventative use only

   b. Sanitation

   c. Certified seed

The experimental basis for these recommendations is extremely limited. Staub and Sozzi (1983) demonstrated some delaying effect of mixtures. However, the magnitude of the delay in field conditions is not known. Milgroom and Fry (1988) did an extensive simulation analysis of these management strategies

using a computer model for metalaxyl resistance in P. infestans. The results of this study generally support Urech and Staub's (1985) recommendations (soil applications not tested with the model). Post-infection use actually resulted in a lower frequency of resistance but greater disease levels than preinfection use. This example is highlighted to show that preinfection use is advisable for disease control in general; its value to resistance management may be negligible if resistance is already present in the population.

One additional controversy that has emerged related to metalaxyl resistance in P. infestans is D.R. Mackenzie's suggestion (1981, Plant Dis. 65:394-399) never to use metalaxyl in potato seed production. The alternative view is to use metalaxyl intensively in seed production to keep populations so small that mutations to resistance will occur only with an extremely small probability. Recent modeling efforts have addressed this question (Milgroom 1990). Model results suggest that when the initial inoculum density is very small, intensive use of metalaxyl in seed production may not result in unacceptable risks of resistance development (Milgroom 1990).

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VIII. FUNGICIDE RESISTANCE IN TOBACCO PRODUCTION

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Major Diseases & Applications

Fungicide use on tobacco in the United States was mainly limited to seed beds until 1980. Weekly applications were made with dithiocarbamates for control of downy mildew (blue mold), damping off, and anthracnose. Since the labeling of metalaxyl on tobacco in 1980, fungicide use has also become widespread in production fields. Currently, more than 50% of tobacco fields are treated preplant with from 0.25 to 3.0 lbs of metalaxyl for the control of downy mildew (Peronospora tabacina), black shank (Phytophthora parasistica), and Pythium soft rot (Pythium spp.). Dithiocarbamate use in the seed beds has declined markedly, now that blue mold is being controlled through use of metalaxyl. Weekly, insurance applications in the seed beds are no longer necessary; instead, fungicides are applied mostly as needed when the disease is active. These practices have resulted in metalaxyl alone being used on tobacco seed beds and fields; increasing the probability that a fungicide resistance problems will develop.

Resistance Monitoring

Monitoring for resistance to metalaxyl in tobacco pathogens is performed regularly by Ciba-Geigy. In addition, Siegel and Nesmith in KY regularly monitor local, national and international populations of P. tabacina for metalaxyl resistance. North Carolina scientists have established base-line data on populations of both the black shank and blue mold pathogens in their state. More work is needed in this area, especially during the growing season. However, adequate safeguards are not available to confine the airborne downy mildews, so studies are not performed during the growing season. Access to USDA facilities at Ft. Detrick have been helpful to date, but increased public access to such a facility would be valuable in the assessment of fungicide resistance and evaluation of control strategies related to fungicide resistance.

Resistance Incidence

Resistance to metalaxyl has not been confirmed for any tobacco pathogen isolated from commercial tobacco production areas of the US. Leaching and poor application have been identified as the reasons for sporadic field failures of metalaxyl.

Resistance Management Strategies

The main management strategy used in the U.S. to reduce the potential for developing resistance to metalaxyl in downy mildew is soil application rather

than foliar applications. Due to its solubility in soil and highly systemic nature, high concentrations of metalaxyl can be placed into all parts of the plant through soil applications, providing control for root, stem and leaf diseases. This may reduce the probability that intermediate-level isolates are allowed to develop and drift the population into resistance.

Metalaxyl is not labeled for foliar use on tobacco in the U.S.; although limited foliar use, in mixture with dithiocarbamates, did occur under emergency exemption status in 1980 and 1981. Concern that such a use pattern could encourage resistance, in part, was responsible for the absence of labeling for foliar use. The logic used was as follows: Tobacco is a very difficult target to cover with foliar materials. Foliar applications of other pesticides used on tobacco do not require complete coverage of the foliage after canopy cover has developed, so producers had neither the equipment nor experience to apply foliar fungicides. Poor coverage of tobacco with a tank mixture of dithiocarbamates plus metalaxyl could have resulted in a possible ideal situation for metalaxyl resistance to develop. For example, some areas of the leaf would have obtained inhibitory concentrations of both fungicides, while others had well below efficacious levels of the protectant and marginal levels of metalaxyl, obtained via the systemic action of metalaxyl. Having areas of the leaf with low levels of metalaxyl without a protectant would appear to be ideal sites for selecting metalaxyl sensitive isolates.

Management of off-label uses may be important in resistance strategies, especially in the absence of strong enforcement to adherence of the label. It is important to acknowledge that some foliar application is made each year during isolated outbreaks of blue mold, despite label restrictions to this use. Control of the current strains can be obtained with as little as 1 or 2 oz/A ai applied at the time of primary inoculum arrival, yet labeled applications to the soil require 32 oz/A much earlier in the season. There is great concern that these low rates will lead to the appearance of resistant tobacco blue mold by encouraging intermediately sensitive isolates.

The current labeling of metalaxyl - using high rates early in the season in order to have adequate concentrations in the plant all season - may have eroded grower confidence in the system and subsequently encouraged off-labeled uses of the product closer to the time of outbreaks of the disease. It should be recognized that producers will not sit back and allow major crop loss to occur when a chemical like metalaxyl is present in the market place; especially with foliar uses on other crops and even on tobacco elsewhere in the world. There is considerable economic savings for the grower to operate a 'wait and see if needed' program. In today's truly international world, there is greater need for common international labeling and enforcement of labeling. Such a program could aid in the management of pesticide resistance and retain uses of valuable chemicals.

An additional factor to consider with tobacco is that an acceptable mixing partner is not present to combine with metalaxyl. The ideal mixing partner should be equally systemic and control the same range of pathogens, so that both the primary and reserve compounds are at the same site all the time. The only acceptable protectant-type compound, from the efficacy standpoint, is mancozeb, but it is not acceptable to the leaf industry. Most tobacco

companies are on record as strongly opposing the use of mancozeb on U.S. grown leaf because of export market concerns.

Metalaxyl is coupled with resistant varieties for black shank control. This strategy is necessary because adequate control does not occur with the chemical alone. It is assumed that this strategy will reduce the development of metalaxyl-resistant strains of pathogen. Similarly, tobacco varieties may soon be available with moderate levels of resistance to blue mold. It may still be necessary to use metalaxyl with these hybrids early in the season prior to the expression of adequate host resistance; a strategy with potential to reduce pathogen resistance to both host plant and chemical.

Prediction of Resistance

The North American Blue Mold Warning System is in place and functions well. This system will alert the tobacco industry to any changes in the status of sensitivity to metalaxyl.

IX. FUNGICIDE RESISTANCE IN TURFGRASS MANAGEMENT

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INTRODUCTION

Many acres of turfgrass are maintained for many purposes in the United States. Turf is grown on golf courses, recreational and sports areas, home lawns and landscaped areas, and for utility purposes, such as roadsides, parks, and cemeteries. The level of maintenance on these turfs varies with use, with golf and other sports turfs receiving the highest levels of maintenance. Golf greens represent the U.S. area of most intensive fungicide use, and the dollar market for fungicides on turf is the largest for any single crop. This heavy use undoubtedly accounts for the fact that most of the instances of fungicide resistance in the U.S. have first been reported on golf turf.

MAJOR DISEASES

A. DOLLAR SPOT - *Sclerotinia homoeocarpa* (*Lanzia* sp. and *Moellerodiscus* sp.)

I. CHEMICALS USED

a. Multi-site inhibitors
anilazine (Dyrene)
chlorothalonil (Daconil 2787)

b. Benzimidazoles
benomyl (Tersan 1991)
thiophanate ethyl (CL 3336, Bromosan)
thiophanate methyl (Fungo 50, Spot Kleen)

1) First reported field control failure (US)

Warren, Sanders, & Cole. 1974. *Sclerotinia homoeocarpa* tolerance to benzimidazole-configuration fungicides. Phytopathology 64:1139-1142.

b) Confirmed field control failures

RI (Jackson), NJ (Clarke), MD (Dernoeden), NC (Lucas), IN (Scott), OH (Shane), MN (Steinstra), MI (Vargas), WI (Worf), PA (Sanders)

All of the above control failures have occurred in grower use situations.

c. Dicarboximides

iprodione (Chipco 26019)
vinclozolin (Vorlan)

1) First reported field control failure (US)

Detweiler and Vargas. 1982. Resistance of *Sclerotinia homoeocarpa* to iprodione. Phytopathology 72:976.

2) Confirmed field control failures

WI (Worf), NJ (Clarke), PA (Sanders), MI (Vargas), MD (Dernoeden), RI (Jackson)

Control failures in WI, NJ, and PA occurred on research areas. Resistant isolates recovered from these areas in WI and PA showed decreased fitness.

3) Iprodione/benomyl control failure

Detweiler, Vargas, and Danneberger. 1983.

Resistance of *Sclerotinia homoeocarpa* to iprodione and benomyl. Plant Disease 67:627-630.

4) Iprodione/chlorothalonil resistant isolates

Pernucci and Jackson. 1983. Tolerance of *Sclerotinia homoeocarpa* (Bennett) to iprodione and chlorothalonil. Phytopathology 73:372.

d. DMI's

fenarimol (Rubigan)
propiconazole (Banner)
triadimefon (Bayleton)

2. NO RESISTANCE MONITORING PROCEDURES IN USE

3. RESISTANCE MANAGEMENT STRATEGIES

a. Because of widespread resistance to benzimidazole configuration fungicides, these chemicals are little used to control dollar spot.

b. Multi-site inhibitors, dicarboximides, and DMI's are alternated. There is no research basis for this strategy.

4. POPULATION DYNAMICS

a. Resistance in S.homoeocarpa to benzimidazoles appears to be stable in control failure sites.

b. Attempts to alter these stable populations failed

Sanders, Gilbride, and Cole. 1982. The effect of single and combination applications of benomyl and chlorothalonil on the level of benomyl resistance in a fungicide modified population of Sclerotinia homoeocarpa. Phytopathology 72:261.

Sanders, Gilbride, and King. 1982. Stability of benomyl sensitive isolates introduced into a benomyl resistant population of Sclerotinia homoeocarpa. Phytopathology 72:265.

Sanders. 1983. Stability of benomyl sensitive isolates introduced into a benomyl resistant population of Sclerotinia homoeocarpa, year 2. Phytopathology 73:374.

B. PYTHIUM BLIGHT - Pythium aphanidermatum

1. CHEMICALS USED

a. Multi-site inhibitors
chloroneb (Terraneb SP, Teramec SP)
etradiazole (Koban)

b. Phenylamides
metalaxyl (Subdue, Pace)

1) First reported field control failures (US)

Sanders. 1984. Failure of metalaxyl to control Pythium blight on turfgrass in Pennsylvania. Plant Disease 68:776-777.

Sanders. 1987. Failure of metalaxyl to control Pythium blight on Kentucky golf courses. Phytopathology 77:121.

2) Confirmed field control failures

IN (Sanders), MD (Sanders), NJ (Clarke),
OH (Shane), TX (Colbaugh)

All of these control failures have occurred in grower use situations

c. <u>fosetyl-aluminum (Aliette)</u>

No reported field failures of Pythium blight control or field isolates of <u>Pythium aphanidermatum</u> recovered showing decreased sensitivity to fosetyl-aluminum.

Sanders & Coffey (manuscript in preparation). Laboratory (MNNG) mutants of metalaxyl-resistant parent isolates of <u>P. aphanidermatum</u> were resistant to metalaxyl and fosetyl-aluminum in vitro and in vivo. Selected double-resistant mutants were highly virulent on fungicide-treated pot-grown perennial ryegrass. These findings suggest that resistance in <u>P. aphanidermatum</u> to fosetyl-aluminum is within the realm of possibility.

d. <u>propamocarb (Banol)</u>

No reported field failures of Pythium blight control or field isolates of <u>P. aphanidermatum</u> recovered showing decreased sensitivity to propamocarb.

2. <u>NO RESISTANCE MONITORING PROCEDURES IN USE</u>

3. <u>RESISTANCE MANAGEMENT STRATEGIES</u>

Sanders, et al. 1985. Reduced rate fungicide mixtures to delay fungicide resistance and to control selected turfgrass diseases. Plant Disease 69:939-943.

Sanders (unpublished). Alternations were effective in resistance stabilization for eight cycles in an experimental population of <u>P. aphanidermatum</u> where the metalaxyl-resistant component showed decreased fitness, relative to the sensitive component.

4. <u>POPULATION DYNAMICS</u>

Sanders & Soika. 1988. Metalaxyl resistance frequency in overwintering populations of <u>Pythium aphanidermatum</u> from metalaxyl control failure sites. Phytopathology 78:1510.

<u>MISCELLANEOUS DISEASES</u>

A. ANTHRACNOSE - <u>Colletotrichum graminicola</u>

1. <u>Confirmed field control failure of benzimidazoles</u>

OH (Shane), MI (Vargas)

Detweiler, Vargas, and Berndt. 1989. Resistance of *Colletotrichum graminicola* to benomyl. Proceedings of the 6th International Turfgrass Research Conference

2. Field control failure of triadimefon (Bayleton)
   WI (Worf)

   This control failure occurred on a research area. Greenhouse confirmation studies are in progress.

B. PINK SNOW MOLD - *Fusarium nivale* (*Microdochium nivalis*, *Gerlachia nivalis*)

1. Confirmed field control failure of iprodione

   Chastagner and Vassey. 1982. Occurrence of iprodione-tolerant *Fusarium nivale* under field conditions. Plant Disease 66:112-114.

C. RED THREAD - *Laetisaria fuciformis*

1. Confirmed field control failure of chlorothalonil
   NJ (Clarke)

   This control failure occurred on a research area.

X. FUNGICIDE RESISTANCE IN VEGETABLE PRODUCTION

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I. CRUCIFERS

Large acreages of broccoli and cauliflower are grown in CA, AZ, and NY.

A. DOWNY MILDEW (*Peronospora parasitica*)

1. Chemicals used for control

a. Multi-site inhibitors
mancozeb
maneb
chlorothalonil
copper

b. Single-site inhibitors
metalaxyl
metalaxyl + mancozeb
metalaxyl + chlorothalonil

2. Resistance incidence

In 1985, resistance in *P. parasitica* to metalaxyl was seen on greenhouse-grown cole crops in CA. Additional problems were seen in cole crop plant beds in GA and CA in 1987.

3. Resistance management strategies: (The uncertainties associated with mancozeb registration status in addition to availability of this chemical make this strategy questionable.)

a. Ciba-Geigy has marketed pre-pack mixtures of metalaxyl with mancozeb and with chlorothalonil to delay the emergence of resistance in *P. parasitica* populations.

b. Seed companies have developed cultivars with tolerance to downy mildew. Where these cultivars are grown, the need for fungicides for control may be eliminated.

II. CUCURBITS (CUCUMBERS AND SQUASH)

Large acreages of cucumber are grown in MI, WI, NC, SC, and TX.

A. DOWNY MILDEW (Pseudoperonospora cubensis)

1. Chemicals used for control

   a. Multi-site inhibitors
      mancozeb
      maneb
      copper
      chlorothalonil
      metiram

   b. Single-site inhibitors
      metalaxyl
      metalaxyl + chlorothalonil
      metalaxyl + mancozeb

2. Resistance incidence

   In 1983, resistance to metalaxyl was detected on cucumber and squash in Florida (Moss, 1987, Plant Disease 71:1045). The following year, it was also detected in the Ciba-Geigy research plots at Vero Beach.

3. Resistance management strategies: (The uncertainties associated with mancozeb registration status in addition to availability of this chemical make this strategy questionable.)

   a. Ciba-Geigy has marketed pre-pack mixtures of metalaxyl with mancozeb and with chlorothalonil to decrease the selection of metalaxyl-resistant populations of P. cubensis.

   b. There are a number of cucumber cultivars with resistance to downy mildew.

B. POWDERY MILDEW (Sphaerotheca fuligenea and Erysiphe cichoracearum)

1. Fungicides used for control

   a. Sulfur
      Sulfur provides fairly effective control of cucurbit powdery mildew, but some cucurbits, such as melons, will not tolerate applications of sulfur.

   b. Single-site inhibitors
      benzimidazoles
      SBI's

2. Resistance incidence

a. Benomyl

1) First report of resistance
Schroeder and Providenti (Plant Disease Reporter 53:271-275, 1969) reported resistance to benomyl in powdery mildew of cucurbits.

2) Further reports of control failures
Benomyl is ineffective for control of cucurbit powdery mildew in CA (Paulus) and NJ (Johnston).

b. SBI's

1) There are reports of reduced efficacy of triadimefon for control of _S_. _fuliginea_-incited cucurbit powdery mildew in CA (Paulus), NJ (Johnston), and FL.

2) Mobay Corporation states on their 1989 label that triadimefon will not provide commercially acceptable control of _S_. _fuliginea_.

3. Resistance management strategies

a. Triadimefon + benomyl in tank mixture provide fairly effective control of cucurbit powdery mildew under CA conditions (Paulus). This strategy, however, would be unwise in benomyl control failure sites, where disease control would accrue from the triadimefon component alone.

b. Cucumber cultivars resistant to powdery mildew are available from many seed companies. Mildew-resistant cultivars of squash and melons are variable with regard to disease resistance in the field.

III. LETTUCE

Large acreages of lettuce are grown in CA, FL, and AZ.

A. DOWNY MILDEW (_Bremia_ _lactucae_)

1. Fungicides used for control
(Maneb is not currently a viable candidate due to the questionable registration status. Aliette is the only fungicide of note available for the control of downy mildew on lettuce).

a. Multi-site inhibitors

copper (Somewhat effective, but may cause yellow leaves with necrotic spots when applied more than once).

maneb

b. Single-site inhibitors

metalaxyl

metalaxyl + maneb

2. Resistance incidence

Metalaxyl was registered for use on lettuce in CA in 1986. Shortly thereafter, with repeated use, this fungicide was ineffective for control of lettuce downy mildew.

3. Resistance management strategies

Following the above resistance development, the metalaxyl label was changed to state that the chemical must be used with a maneb fungicide at full label rates for lettuce downy mildew control. Again, this strategy should not be employed in sites where a metalaxyl control failure is known to have occurred.

B. LETTUCE DROP (*Sclerotinia minor*)

Trials of fungicides for control of lettuce drop in the Salinas Valley (CA), 1985-1988, have produced variable results with respect to the presence of dicarboximide resistance.

In 1985, in one of three trials, vinclozolin-treated areas were not significantly different from untreated. Dicloran alone or dicloran + vinclozolin provided excellent control of lettuce drop in the same test. In 1986, in one of three trials, iprodione treated areas were not significantly different from untreated. Dicloran + vinclozolin provided excellent control. In 1987, in two of four trials, iprodione treated areas were not significantly different from untreated. Dicloran + iprodione provided excellent control of *Sclerotinia minor*. In 1988, all fungicide treatments were significantly better than the untreated controls.

No attempt was made to confirm by isolation whether these apparent dicarboximide control failures were resistance-related.

XI. FUNGICIDE RESISTANCE IN STRAWBERRY PRODUCTION

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Disease description and materials available for control:

Strawberries are subject to attack by a number of pathogens, however *Botrytis cinerea* (Gray Mold) seems to be the most prone to develop resistance to fungicides for several reasons: 1) it is able to infect during the entire strawberry blossom and production cycle, thus, it is exposed to (selected by) control agents for an extended period of time; 2) it sporulates very prolifically, increasing the likelihood of the occurrence of a resistant genotype; 3) it has a rapid generation time, therefore, multiple generations are subjected to selection in a single season; and 4) it is multinucleate, lending a greater degree of genetic diversity. The threat of resistance development is far greater in California production areas, at least in part, because of their extended harvest and "ever-bearing" varieties.

Several fungicides have been used to control Gray Mold with varying degrees of success. Thiram is moderately active against *Botrytis* and other pathogens and is sometimes used in combination with other fungicides. The visible surface residue it leaves, however, can reduce the value of the crop. Captan also has multiple pathogen activity and has effectively controlled Gray Mold when disease pressure is light. It was a common tank mix companion until the recent extension of the pre-harvest interval to four days limited its use during the picking season. Benzimidazoles (benomyl and thiophanate-methyl) were very active against *Botrytis* until their utility for Gray Mold control became severely limited due to widespread resistance. Vinclozolin is the only dicarboximide fungicide currently registered to control Gray Mold on strawberries, however, iprodione is registered for *Botrytis* control on several other crops. Both fungicides are extremely effective in Gray Mold control. Development of resistance has occurred and is discussed below.

Monitoring programs and methods and resistance status:

Dicarboximide use on strawberries in the U.S. began in 1981. Because of resistance development in *Botrytis* populations on grapes in Europe, monitoring began immediately. These programs were not systematic, however, and were intended only to provide an early warning if resistant strains were detected. From 1981 to 1983, most monitoring was done in the Watsonville area of CA. Results from 1983 monitoring indicated that resistant isolates comprised <0.01% of the population. In 1984, this level increased. No disease control failures occurred, however, it was noted that disease control provided by vinclozolin was reduced. All strawberry production areas in CA were sampled in 1985. Although dicarboximide-resistant strains of *Botrytis* were detected in all regions, they were most prevalent in the Watsonville area. Several of the monitoring programs also tested for benzimidazole and captan resistance. *Botrytis* strains that were resistant to one, a combination of two, or all three fungicides were detected. Monitoring from 1986 to the present indicates

that dicarboximide resistance is present in all strawberry production areas of CA. Monitoring programs in NC, NY, FL, and LA have not detected resistant strains, however, one resistance report has come from NY. In a recent monitoring survey of strawberries grown in CA and purchased in NC markets, 95% of recovered Botrytis isolates were benzimidazole resistant and 22% were resistant to dicarboximides.

The most commonly used monitoring technique for Botrytis resistance to dicarboximides is a process of isolation of the fungus from selected rotted strawberries into pure culture followed by transfer to medium amended with 10 ppm dicarboximide. This method, though useful, is labor intensive and requires 5 to 10 days to obtain results. A recently developed method involves the direct plating of spores from sporulating lesions onto a semi-selective, indicator medium. Sampling is greatly simplified, and results are available in 24 to 36 hours. This method was implemented in 1988 and will greatly aid the monitoring process.

Unlike benzimidazole resistance, which tends to be stable, the frequency of dicarboximide resistance in Botrytis populations will decline during periods of nonuse of dicarboximides. This trend has been observed, although detailed studies of population dynamics have been confounded by the mobile nature of the airborne Botrytis spores. Additionally, vinclozolin continues to provide a degree of disease control in the presence of certain levels of resistance frequency, which allows the fungicide to be useful in an appropriate disease control program.

Resistant management strategies for dicarboximides:

In 1985, BASF Corporation restricted the number of vinclozolin applications permissible on strawberries. Since resistance had been detected in CA the number of applications per season in CA was limited to four. In other states where the resistance threat did not appear to be as great, six applications were permitted. In addition, a statement was appended to the label warning of the possibility of resistance. It further suggests that alternation with other fungicides might reduce the likelihood of resistance. The label also recommends that if loss of disease control due to resistance occurred, a fungicide with a different mode of action should be used.

Vinclozolin is currently the only dicarboximide fungicide registered for disease control on strawberries in the U.S. Iprodione, however, is registered on other crops for Botrytis control, and may soon be registered on strawberries. As part of an overall dicarboximide management strategy, in 1988, BASF Corporation and Rhone-Poulenc Ag Company have agreed to match the restrictions of the number of permissible dicarboximide applications on all crops with common registration. In addition, both the vinclozolin and iprodione labels will bear a statement to prevent increasing the total number of dicarboximide applications by using two different fungicides e.g., if two applications of one dicarboximide fungicide are applied to strawberries in CA, only two applications (not four) of the other dicarboximide may be applied, thus abiding to the limit of a total of four dicarboximide applications.

These strategies have been partially successful. As noted above, however, alternatives to dicarboximide fungicides may also be subject to resistance or have other undesirable properties. Prudent use of dicarboximide fungicides will help preserve and prolong their usefulness. The development and careful integration of fungicides with different modes of action are essential in resistance management and effective disease control.

XII. FUNGICIDE RESISTANCE IN MUSHROOM PRODUCTION

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Introduction

This specialty crop is grown in 28 states, and during crop year 87/88, was worth $544.2 million at farm doors across the United States. In Pennsylvania, the mushroom crop is valued at more than the combined value of potatoes, fruits, and vegetables ($194.5 million vs. $134 million, 86/87). Importation of processed mushrooms from Pacific rim countries during the past 20 years has resulted in U.S. mushroom growers redirecting their production to fresh market outlets (25% in 1971/2 vs. 65% in 1987/8). This market shift has required development and adoption of new growing techniques and germplasm. Dependence on the fresh market requires that mushroom farmers grow, package, and deliver appealing, blemish-free mushrooms to retailers, restaurants, and institutional buyers.

Major diseases

I. Verticillium brown spot (*Verticillium fungicola*) is the most frequently encountered disease at mushroom farms, and its occurrence constitutes a serious problem on mushrooms being grown for the fresh market. The disease tends to be endemic, irrespective of the time of the year. A 1980 survey of 35 mushroom crops reflected disease occurrence in every crop surveyed with an increasing incidence as the crop aged, from .01% on first break to 20+% by fourth break. A qualitative survey of disease occurrence in 1984 yielded data from 60% of Pennsylvania mushroom farms, and listed brown spot as the most significant disease for each crop.

a. Chemical Control

Chemicals registered to control this disease include dithiocarbamate, zineb, and the benzimidazoles (benomyl and thiabendazole).

1) Resistance monitoring has consisted of bi-yearly resistance assessments (since 1978) using media amended with benomyl, thiabendazole, or both. Isolates of the pathogen are obtained from farms across the U.S. with isolation and resistance assessments being made at Penn State.

b. Resistance Incidence

1) Benomyl has not been effective in inhibiting germination of conidia or vegetative growth of the fungus in 95% of the isolates tested. During the 1980's, there tended to be more isolates which grew at 50 ppm benomyl than there were in the late 1970's. This

monitoring procedure was initiated in about 1973, when mushroom farmers throughout the U.S. reported lack of disease control with benomyl.

2) Isolates of *V. fungicola* respond somewhat differently to thiabendazole, in that laboratory assessments result in data akin to that for benomyl, yet disease occurrence and increase are reduced by the chemical in controlled cropping experiments. Thiabendazole decreases the total number of Verticillium-spotted mushrooms by 15-35%. Farmers have not corroborated these data as they cannot perceive varying disease levels when Verticillium brown spot is less than completely controlled. Further complicating this situation is the belief by farmers that one of the wetting agents in the flowable thiabendazole formulation enhances the likelihood of bacterial blotch, another quality-reducing disease.

c. Resistance Management

There are no mushroom cultivars with resistance to Verticillium brown spot, so this option is not available. Modifications to cultural practices have been under investigation for five years, and some disease reduction may be possible with this approach. The need for sanitation and hygiene have only recently been emphasized, and their implementation at mushroom farms is still in the developmental stage. Another strategy, the routine use of zineb, while reserving benomyl for occasional use, has not resulted in the reemergence of benomyl-sensitive strains as the predominant phenotypes at mushroom farms.

1) The import license for zineb was suspended by Department of Commerce in 1988, so supplies of zineb in distribution lines are almost nonexistent.

2) Chlorothalonil was reregistered in summer, 1988, and although IR-4 has reactivated their request for a mushroom tolerance, neither the tolerance nor a label are available for this broad spectrum fungicide on mushrooms. Reports from Europe, Oceanasia, and England indicate that chlorothalonil has been minimally effective during its 10-12 years of usage at mushroom farms in these countries. Currently, CA and TN have Section 18 labels for the use of chlorothalonil on mushrooms.

d. Availability of research on fungicide resistance in V. fungicola

There are no current studies as there are few options in chemical disease control. Thus, resistance management, population shifts, and mechanisms of resistance are academic questions with no potential of benefiting mushroom growers.

II. La France (virus induced)

This disease is a significant crop-reducing disease, and management is aimed at sanitation, hygiene, and indexing. Use of zineb dust (15%) as a protectant has been effective in minimizing secondary infections by basidiospores from La France diseased mushrooms. There is no alternative to zineb, and resistance to zineb in *Agaricus bisporus* basidiospores has not been reported.

III. Mildew Diseases

The pathogens responsible for these diseases are *Trichoderma viride* and *Hypomyces roseullus* (*Dactylium dendroides*, imperfect stage). These diseases can be significant when they occur, but their occurrence is infrequent when compared with Verticillium brown spot. There are no research reports of resistance in either of these pathogens to benzimidazoles. Anecdotal information from farmers suggests that *H. roseullus* is resistant to benomyl, however, these reports have not been validated through research.

XIII. FUNGICIDE RESISTANCE AS IT RELATES TO EPA POLICIES AND PRIORITIES

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EPA's concern and attention to fungal resistance to pesticides and fungicides, in particular, has become a routine and integral part of all phases of its regulatory function. There are six specific areas of the Agency's function where resistance to fungicides play a role. These areas are:

1. The registration system
2. The emergency exemption practice
3. The registration standards procedure
4. The special review practice
5. The Data Call-In process
6. Extramural activities

Registration System

For the registration process, resistance to fungicides comes into play in the label review. Resistance became a critical issue when resistance to benomyl and thiophanate-methyl first became apparent. At that time labeling policy and acceptable language were worked out through negotiations with the registrants. The resulting policy can be summarized by the following:

1. Registrant must specify that a resistance problem exists.

2. Registrant must state that if a chemical is not effective, it may be due to the existence of resistant strains and that use of the product must be discontinued.

3. If cross-resistance problems are known to exist, guidance on which other fungicides are to be avoided as alternatives must be stated.

4. In the matter of delaying the onset of resistance, the registrant cannot recommend the use of tank mixes to the exclusion of alternate applications. For example, a combination of benomyl and maneb cannot be recommended over alternating benomyl with maneb, unless there is conclusive data that tank mix is more efficacious than alternate applications.

Emergency Exemptions

An emergency exemption known as a Section 8 authorizes the EPA Administrator to exempt state agencies from compliance with registration limitations. The exemption is granted, providing an emergency situation exists, and significant economic loss can be expected if the emergency situation is not addressed. Emergency conditions hinge on the following:

1. No effective pesticide or no effective cultural control practice is available.

2. The pest is new to the area.

A valid reason for granting an emergency exemption based on unavailability of effective chemicals is the loss of effectiveness of available chemicals due to resistance. As an indication of how resistance to fungicides enters into the emergency exemption process, the following figures are of interest. Based on an analysis of four years of requests for exemptions, the following applies to fungicides.

1. An average of seventeen requests were made per year.

2. Of these seventeen requests, twelve were based on claims of ineffective alternatives.

3. Of the twelve, three were based on loss of effectiveness due to resistance. The three requests were granted.

The Registration Standard Procedure

The Registration Standard Procedure is a review of the available information that the Agency has in its files on a specific active ingredient pesticide. It includes examination of data on product chemistry, regulatory status, and use information.

The use information is a summarization of two reports; the Qualitative Use Assessment and the Quantitative Use Assessment. The Quantitative Use Assessment addresses factors such as volumes of the pesticide produced and quantities used at the crop site. The Qualitative Use Assessment identifies the use sites, application rates, formulations, and methods of application. Specifically, the report does address whether or not a fungicide is subject to resistance and/or how it may be used to mitigate development of resistance.

To round off the story on the Registration Standard Procedure: If, at the end of the review, it is determined that necessary data is unavailable, the Agency requires that the registrants supply such data within a certain time period.

Data Call-In

Data Call-In letters also known as 3(c) 2(b) letters may be the outcome of the Registration Standard Procedure. In a data call-in, the registrants are required within a specific time period to supply data which is missing or which is critical in determining risk or benefit of a chemical. On the risk side, it is often required that toxicity or residue data be supplied. On the benefits side, the requirement may deal with information on percentage of crop treated, comparative performance efficacy data. In so far as fungal resistance is concerned, the registrant may be required to supply a report on the potential for resistance to the chemical concern and to chemicals which are alternative control measures.

The Special Review Process

Often, the findings of the Registration Standard Procedure or the acquisition of new data on toxicity or environmental effects will trigger the Special Review Process of a chemical. In this complex process, the risks and benefits are examined. Usually, various regulatory options for reducing risks are proposed, (e.g., requirement for more stringent precautionary measures for applications or cancellation of registrations for certain high risk dietary exposure sites of application).

The Special Review Process demands an indepth examination of benefits as well as risks. The benefits summation document is presented in the Benefits Analysis for each crop for which the chemical is registered for use. Benefit Analysis addresses:

1. Registration Summary

    a. Diseases controlled
    b. Methods of application
    c. No. of applications
    d. Formulations used
    e. Chemical alternatives

2. State recommendation

3. Outlook for new chemicals

4. Non-chemical control measures

5. Discussion of specific diseases controlled

6. Comparative Performance Evaluation

7. Economic impact of the loss of a registration or restricted use

In discussing alternative chemicals, fungal resistance is a guiding factor in determining what is referred to as the viable alternative chemicals. Viable alternatives are those which are economically feasible, non-phytotoxic, not prone to resistance, and comparatively efficacious.

Promoting the Management of Resistance

To illustrate the high priority EPA attaches to pesticide resistance, the Agency is involved in the promotion of an international pest resistance management project. One Agency plant pathologist is involved almost full time in the project. The Agricultural Research Institute is sponsoring an International Congress implementing global pest resistance management strategies. A planning committee has been formed to develop and conduct the Congress and facilitate follow-up activities. The planning committee is currently comprised of representatives of the USDA, EPA, The Agency for International Development, The National Audubon Society, The National

Agricultural Chemical Assoc., The World Bank, and the agrochemical industry. The planning committee recognizes that without intervention, pest resistance may eventually limit the efficacy of most plant protection chemicals.

The Congress will be preceded by efforts of working groups; the contributions of the working groups will be reviewed and finalized at a Plenary Working Group. The consensus reports will be distributed to those invited to the Congress. The Congress will make recommendations and hopefully promote implementation of the recommendations. The working groups will begin functioning in late 1989, and the Congress will be convened in September of 1991.

CONCLUSIONS AND RECOMMENDATIONS

A. FUNGICIDE RESISTANCE IN THE US

It is clear from the information synthesized and contributed by this panel of experts, that resistance to systemic fungicides in the United States is present and increasing as the use of these chemicals proliferates. This is alarming, since it underscores the fact that all of the currently registered systemic fungicides are in jeopardy, as well as those yet-to-be-registered compounds that are in the same chemical classes as those systemics presently used in U.S. agriculture. Individual summaries in this report have detailed control failures due to resistance in the following groups of chemicals:

1. Benzimidazoles

    scab (*Venturia inaequalis*) - apples
    blue mold (*Penicillium expansum*) - apples, ornamentals
    scab (*Elsinoe fawcetti*) - citrus
    greasy spot (*Mycosphaerella citri*) - citrus
    green mold (*Penicillium digitatum*) - citrus
    gray mold (*Botrytis cinerea*) - grapes, ornamentals, strawberries, cherries
    powdery mildew (*Uncinula necator*) - grapes
    dollar spot (*Sclerotinia homoeocarpa*) - turfgrasses
    anthracnose (*Colletotrichum graminicola*) - turfgrasses
    early leafspot (*Cercospora arachidicola*) - peanuts
    late leafspot (*Cercosporidium personatum*) - peanuts
    brown blossom blight/fruit rot (*Monilinia laxa*, *M. fructicola*) - apricots, peaches, nectarines, cherries, almonds
    scab (*Cladosporium carpophilum*) - peaches, nectarines
    green fruit rot (*Monilinia* spp, *Botrytis cinerea*) - peaches, cherries
    brown spot (*Verticillium fungicola*) - mushrooms

2. Dicarboximides

    gray mold (*Botrytis cinerea*) - ornamentals, strawberries
    dollar spot (*Sclerotinia homoeocarpa*) - turfgrasses
    snow mold (*Microdochium nivalis*) - turfgrasses
    Sclerotinia blight (*Sclerotinia minor*) - peanuts (resistant isolates only)
    lettuce drop (*Sclerotinia minor*) - lettuce

3. Benzimidazoles and dicarboximides (multiple resistance)

    gray mold (*Botrytis cinerea*) - ornamentals
    dollar spot (*Sclerotinia homoeocarpa*) - turfgrasses

4. Sterol biosynthesis inhibitors (DMI's)

scab (*Venturia inaequalis*) - apples (reduced sensitivity)
gray mold (*Botrytis cinerea*) - grapes (reduced sensitivity)
anthracnose (*Colletotrichum graminicola*) - turfgrasses (suspected control failure)
powdery mildew (*Uncinula graminicola*)

5. Phenylamides

Pythium blight (*Pythium aphanidermatum*) - turfgrasses
Downy mildew (*Bremia lactucae*) - lettuce
Downy mildew (*Peronospora parasitica*) - cauliflower, broccoli
Downy mildew (*Pseudoperonospora cubensis*) - cucumbers, squash

All of the currently registered systemic fungicides in the U.S. are included in the above listed four groups. Although there are many systemic fungicides registered for use in the U.S., the phenomenon of cross-resistance prescribes that, from the standpoint of fungicide resistance, there are, in fact, only four systemic fungicides (the above four groups) available for use on many crops. On some crops, only the systemic fungicide is available for critical disease control. When viewed from the perspective of fungicide resistance, it is apparent that our arsenal of fungicides is ominously small. It is essential, therefore, that currently registered protectant fungicides, such as anilazine, chlorothalonil, dichloran, and the dithiocarbamates, continue to be available in order to maintain maximum diversity in chemical control options. Protectant fungicides offer, in many host/pathogen systems, the only companion fungicides available for use in resistance management strategies. As such, they are critical components of the available fungicide arsenal.

B. MONITORING AND SCREENING

There is great need for data on baseline sensitivity of pathogen populations prior to exposure to fungicides. Such data are essential for use as a standard against which post-fungicide sensitivity can be compared. While the threat of resistance to systemic fungicides appears to be proportional to their use, there appears to be little systematic monitoring for resistance in pathogen populations that are routinely treated with at-risk fungicides. Also greatly needed is the development of simple, quick, and reliable techniques for monitoring and screening of the multiplicity of host/pathogen systems for resistance in pathogen populations. Directly related to the foregoing is the pressing requirement for containment facilities wherein monitoring and research can be carried out with resistant pathogens without fear of spreading these fungi into uncontaminated areas. Without such safeguards, there is great risk in transporting resistant pathogens or contaminated plant tissue bearing resistant obligate parasites from region to region within the country.

C. RESEARCH AND FUNDING REQUIREMENTS

There is shockingly little research available in the critical areas of (i) resistance risk assessment, (ii) resistance management strategies, and (iii) dynamics of fungal populations with respect to resistance. The absence of

research guidance seriously hampers attempts at sound management of the problem of fungicide resistance. Persons who have responsibility for advising growers on resistance management strategies find themselves in the position of knowing only how <u>not</u> to use systemic fungicides and can only guess what strategies may be effective to delay resistance and prolong the useful lives of these valuable chemicals. Mixtures and alternations of fungicides have been suggested as possible use strategies for avoiding or delaying resistance control failures. These protocols, however, have been little studied, so that scientifically based recommendations are seldom possible.

There is little inducement for researchers to explore critically-needed research areas without significant <u>commitment of funding agencies</u> to addressing the many problems that remain in the area of fungicide resistance. Funding commitment is unlikely to accrue without awareness on the part of funding agencies that the problem of unmanaged fungicide resistance has the potential to seriously threaten U.S. agriculture.